OHIO STATE UNIVERSITY
MAY 29 1957
LIBRARY

BULLETIN ON

NARCOTICS

Vol. VIII, No. 4, OCTOBER-DECEMBER 1956

UNITED NATIONS

Bulletin on NARCOTICS

Vol. VIII, No. 4, OCTOBER-DECEMBER 1956

UNITED NATIONS

EUROPEAN OFFICE

Geneva

NOTE

Reproduction of unsigned material is authorized and welcome. For permission to reproduce signed material, request should be made to the Secretariat of the United Nations. It would be appreciated if a copy of any publication containing material reproduced from the *Bulletin* were sent to the Division of Narcotic Drugs, Palais des Nations, Geneva.

* * *

Opinions expressed in articles are not necessarily those of organs or Members of the United Nations.

Table of Contents

Page

GENERAL

A new plant source of narcotic drugs: Methysticodendron Amesianum, by R. E. Schultes 1

The search for a non-addicting analgesic, by H. Isbell, M.D. 5

Khat 6

TECHNICAL

Experimental evaluation of analgesia, by C. Radouco-Thomas, M.D., S. Radouco-Thomas, and Gl. Nosal 14

Quantitative determination of morphine in opium by paper chromatography and spectrophotometry, by H. Asahina and M. Ono 39

OFFICIAL

11th session of the Commission on Narcotic Drugs 45

22nd session of the Economic and Social Council 47

A New Plant Source of Narcotic Drugs : Methysticodendron Amesianum

By Dr. Richard Evans Schultes, F.L.S.
Curator, Ames Herbarium, Botanical Museum of Harvard University

Recent ethnobotanical investigations in Colombia have greatly added to our knowledge of the narcotic and stimulant plants which are used in divination, witchcraft and medicine by the Indians of these areas. The discovery, in the high and geographically isolated Indian village of Sibundoy, of a tree, representing a new genus and belonging apparently to the *Solanaceae*, may have significant consequences in several different fields of study.

In early 1942, whilst I was engaged in ethnobotanical studies in the Putumayo of Colombia, my attention was drawn to a most extraordinary tree which the Kamsá and Inga Indians of the valley of Sibundoy cultivate for use as a narcotic. Flowering material was collected, an examination of which seemed to indicate that a new genus close to the *Brugmansia* group of *Datura* had been found. In the following years, I made several trips to Sibundoy and was able to gather additional material from the original and several adjacent trees; and colleagues, who have travelled in the area, have been kind enough to prepare specimens from still other trees both in the village of Sibundoy and the neighbouring town of Santiago. In 1953, we were able finally to introduce the tree into cultivation in easily accessible gardens. Now, with plentiful material and after more than a decade of cautious consideration, I still hold that we have at hand an undescribed genus.[1] In 1956, I was able to continue very detailed field studies on the genus in Sibundoy.

Use of the Plant in its Original Habitat

This plant, *Methysticodendron Amesianum*, known only from the valley of Sibundoy, is employed by the Inga and Kamsá Indians in their witchcraft. Its use in each tribe is restricted to several witch-doctors, and these practitioners grow the plant with special care in the neighbourhood of their huts. The wild habitat of the species is said to be the forested slopes of the mountains to the south of the valley of Sibundoy, especially the slopes of the botanically almost completely unknown Cerro Patascoy. When in cultivation, the plant is apparently the hereditary property of certain families, as the sorcerers pass it on to the eldest son together with the secrets attending its use.

Little could be learned about the use of the narcotic, partly because of the proximity of influential Christian missionaries. It would appear from what information I have been able to glean that *Methysticodendron Amesianum*, which the Indians classify together with other intoxicating solanaceous plants similarly used, is by far the strongest of all the narcotics of the region, surpassing in danger and potency even the tree-species of *Datura*.

A description of the intoxication indicates that the active principles may be solanaceous alkaloids of the tropane series. This intoxication, resorted to by the witch-doctors only for very important or difficult cases of divination, prophecy or therapy, usually lasts for two full days and sometimes may persist for four—with a long period of complete lack of consciousness. Missionaries who have been at work in the valley of Sibundoy for more than a quarter of a century suspect that the death of one aged witch-doctor may have been due to an overdose of this narcotic.

FIGURE 1

Habit drawing of a flowering bunch, about one half natural size (Drawn by Elmer W. Smith)

[1] A botanical description of the tree is presented in the appendix.

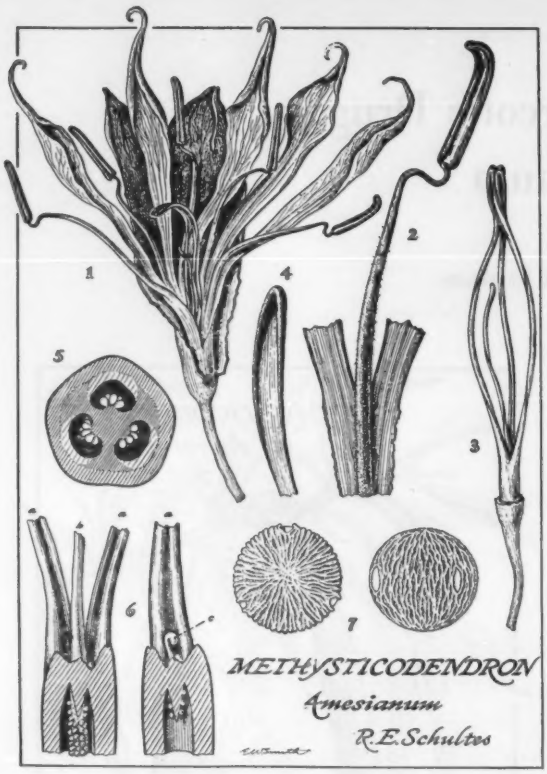


FIGURE 2

1, partly dissected flower, about one half natural size, showing the relative sizes of stamens, styles and other floral segments. 2, a stamen and its attachment, slightly more than natural size. 3, the styles and the style-like appendage, about three quarters natural size. 4, the tip of a style showing the stigmatic surface, about three times natural size. 5, cross section of the ovary, about six times natural size. 6, sagittal section of the apical part of the ovary showing (a) styles, (b) the large appendage homologous with the styles and (c) one of the minute appendages arising from the central tissue of the ovary, about three times natural size. 7, pollen grains, about four hundred and fifty times natural size (Drawn by Elmer W. Smith)

Smaller doses of the drug are administered to boys who are studying witchcraft. Certain of the medicine-men's secrets are apparently imparted only when the novitiate is " under the protection " of this narcotic.

According to the sparse information available, only the leaves of *Methysticodendron Amesianum* can be employed in the preparation of the narcotic infusion. The leaves, which must be gathered not more than one hour before the narcotic drink is to be taken, are stripped from the plant, crushed and allowed to remain in cold water for perhaps one half-hour. Just before the draught is to be taken, the leaves and the infusion are slightly heated and agitated. Then the liquid is strained. It is apparently never allowed to boil.

Witch-doctors never take the whole draught at one time, it is said; but they may drink as much as a large cup-full over a period of two or three hours. If, by then, the desired frenzy and initial narcosis have not been induced, an assistant prepares additional drink from fresh leaves.

According to the Indian tradition, this narcotic must be taken only during the wane of the moon.

In addition to its use in divination and prophecy, *Methysticodendron Amesianum* is employed by its " owners " in the treatment of certain diseases. The leaves and flowers are heated in water and applied in the form of a plaster for the reduction of tumours and swellings, especially of the joints. For persistent chills and fevers (advanced tuberculosis (?)), which are not uncommon at this high, damp altitude, the witch-doctor will sometimes first bathe the whole body of a patient with a warm decoction of the leaves and flowers, then smear lamb-fat on the chest, back and abdomen.

The Kamsá name of *Methysticodendron, mets-kwai borrachero* or *mits-kway borrachero* means " intoxicant of the jaguar ". The term *borrachero* is a loan-word from Spanish, for in southern Colombia and Ecuador a large number of solanaceous narcotics are referred to popularly as *borrachero* (*borracho* = drunken). Notwithstanding the meaning of the Kamsá *mets-kwai*, these Indians refer to the plant in Spanish as *culebra-borrachero* (" intoxicant of the snake ") and not as *tigre-borrachero*. Another name which the Kamsá people apply to the plant is *goon'ssi-an borrachero*, the meaning of which I could never ascertain. The Inga Indians of the valley of Sibundoy sometimes call the plant *kin-de-borrachero*, but recent field work indicates that this is more properly applied to a species of tree *Datura* with abnormal leaves due to virus infection.

FIGURE 3

Kamsá Indian boy holding a flower of Methysticodendron Amesianum, Sibundoy, Colombia (Author's photograph)

Cultivation of the Plant outside its Habitat

Because of the extreme rarity of this plant, even in the valley of Sibundoy, and because of its extraordinary beauty, several attempts were made to introduce *Methysticodendron Amesianum* into cultivation. In 1953, efforts to establish it by planting short sticks cut from the upper branches met with success. The species is now in cultivation in the Capuchin Mission Station at Sibundoy; in the gardens of the Ciudad Universitaria in Bogotá, Colombia; in the Universidad del Cauca, Popayán, Colombia; at the Royal Botanical Gardens, Kew; in the greenhouses of the Department of Biology, Harvard University; at the Division of Plant Exploration and Introduction, Bureau of Plant Industry, Beltsville, Maryland; and at Wongwood Gardens, Kennet Square, Pennsylvania.

Chemical Analysis of the Active Principle

There has as yet been no full chemical analysis of the active principle of *Methysticodendron Amesianum*, but material for this purpose has recently been procured.

FIGURE 4

Habit photograph of a flowering bunch of Methysticodendron Amesianum, Sibundoy, Colombia (Author's photograph)

FIGURE 5

Typical witch doctor, Sibundoy, Colombia (Author's photograph)

APPENDIX

Botanical description and comments

Methysticodendron—*R. E. Schultes* in *Bot. Mus. Leafl. Harvard Univ.* 17 (1955) 2.

Tree. Leaves ligulate. Flowers solitary, very large, hanging. Calyx long-tubular, herbaceous, spathaceous. Corolla very deeply 5-divided, with spathulate, acuminate lobes. Stamens affixed near apex of tube, with rather strong filaments which are attenuate towards apex and strongly twisted; anthers linear, with parallel locules which are introrsely longitudinally dehiscent. Ovules numerous in each locule, axillary. Styles 3, free (but to the eye simple, coherent with mucilage (?), strongly twisted), rather fleshily filiform-ligulate, but in section concave-complanate; stigma undivided, clavellate-tumescent; 2 or 3 appendages (1 filiform, rather similar to style but much shorter and 1 or 2 reduced to very short subulate outgrowths or bullae) arising from central part of ovary. Fruit not present.

Methysticodendron Amesianum—*R. E. Schultes* in *Bot. Mus Leafl. Harvard Univ.* 17 (1955) 2.

Tree up to 25 feet tall, with short, densely leafy branches. Leaves membranaceous, dark green, narrowly linear-ligulate, apically acuminate, basally gradually attenuate (very often without distinct petiole), marginally commonly subundulate or

undulate, in adult state 20–26 cm long, 1.3–2.0 cm wide; central nerve strong, beneath strongly elevated, secondary nerves almost parallel, somewhat conspicuous, towards margin arcuate-anastomosing; both surfaces minutely and irregularly (near veins denser) pilose. Flowers up to 28 (mostly more or less 23) cm long apically 10–13 cm in diameter in bud, at dusk and night very strongly and sweetly fragant. Calyx spathaceous, green, papyraceous or membranaceous, 2- to 5-parted, with acute teeth, tubiform for half its length, 3/5 as long as corolla (up to 13 cm long), both surfaces very minutely and irregularly pilose. Corolla divided for 3/5–4/5 its length, with lobes usually 5 (but abnormally 4 or 6), membranaceous, white, spreading, spathulate or almost spathulately rhombiform, apically long acuminate, circinnate, entire, upper half strongly inflexed, 14–16 cm long, towards apex of tube usually 6 mm. (but in widest part 2.5–3.3 cm) wide, near base very remotely and irregularly pilose within and without, with 3 conspicuous, strongly parallel veins; tube rather densely white-pilose within. Stamens usually 5, shorter than corolla lobes; with terete filaments, more or less 5 cm long, 2 mm in diameter at base, apically strongly twisted and lightly bent, pilose except at apex; anthers yellow, 2.7–3 cm long, 3–4 mm in diameter. Ovary subcylindric, conspicuously 3-sulcate, glabrous, up to 1.5 cm long, 0.5 cm in diameter, gradually produced into styles. Styles glabrous, up to more or less 9 cm long, 2.5 mm in diameter, apically slightly clavellate and strongly flattened, with stigmatic tissue on very apex and along adges up to 1–1.5 cm in decurrent form, black (when dry) and papillose. Appendix (rudimentary style?) like styles arising from central part of ovary, up to 5.5 cm long, accompanied by 1 or 2 subclavellate, fleshy appendices 2–5 mm long.

Methysticodendron is here described as a genus of the *Solanaceae* with some hesitation, for the concept shows several characters which are not easily accommodated in that family.

In habit, *Methysticodendron Amesianum* is a small tree, quite suggestive of some of the larger tree-species of *Datura*—i.e., *D. arborea L., D. candids D. suaveolens RBK.* and *D. sanguinea R. and P.* Like these members of the *Brugmansia* section of *Datura*, the new concept is characterized by large pendulous flowers. The intoxication which a decoction of the leaves of *Methysticodendron Amesianum* induces bespeaks the presence of similar alkaloids : hyoscyamine and scopolamine. There are also strong similarities in the branching, as well as in the structure of the calyx and stamens.

The pollen grains of the type plant of *Methysticodendron* have been studied by Dr. Thomas van der Haman, palynologist to the Servicio Geológico Nacional of Colombia, and by Professor Elso S. Barghoorn of the Botanical Museum of Harvard University. Dr. Barghoorn's description of the grains follows :

> " Pollen grains spherical, chiefly inaperturate, occasionally tricolporate or triporate with very weakly developed short furrows. Size ranging from 45 μ to 63 μ, averaging about 53 μ (after acetylation). Sculpturing of the exine striate, and of the ' finger print ' pattern of striation. Striae converging at the poles and commonly anastomising between the poles. Polar areas reticulate-foveclate. A feeble punctate or reticulate texture may be detected between the striae or along their sides. Equatorial costae conspicuous.
>
> " In basic morphology, the grains are a modified bipolar, tricolpate type, in which both pores and furrows have been greatly reduced. "

The pollen grains of *Methysticodendron* are of the same type and are, indeed, very similar to the grains of other species of *Datura*, section *Brugmansia*.

Notwithstanding these similarities, *Methysticodendron* can be distinguished at once from the tree-species of *Datura* by its very deeply lobose corolla, which is divided for three-fifths to four-fifths of its length. The tubular portion of the corolla is wholly enclosed within the tubular, spathaceous calyx, so that it bears a superficial resemblance to a polypetalous flower, the gamopetalous condition not being evident as the blossoms hang on the tree. The narrowly ligulate leaves are also strikingly unlike those of the tree-species of *Datura*.

The major and perhaps fundamental departure from the solanaceous condition, however, lies in the morphology of the ovary and styles. *Methysticodendron* has three conduplicate carpels with three free styles (with undivided stigmatic areas) which retain the conduplicate condition. The concave structure of the styles leaves the ovule cavity somewhat open at the apex, exposing the apical ovules, until, about one third of the way down, the carpel walls fuse to form a trilocular syncarp. All members of the *Solanaceae* have a simple style with a two-lobed stigma. All members of the tribe *Datureae* have a four-celled ovary. Indeed, the only tribe of the family possessing a three- to five-celled ovary is the *Nicandreae*, but even *Nicandra* has a simple style.

The structures most difficult of interpretation are the several appendages arising from the central tissue of the ovary. One of these is an elongated appendage usually entirely homologous with the styles (even to the presence of what appears to be stigmatic tissue), but much shorter. Accompanying this appendage are usually one or two anomalous knob-shaped, clavate or even subulate projections. These do not appear to arise consistently from any definite part of the ovary. They resemble adventitious stylo-like outgrowths.

Several fruits of *Methysticodendron Amesianum* were collected in 1947. These were preserved in alcohol and sent to Bogotá, but, unfortunately, they were lost in shipment. Consequently, a detailed description of them cannot be given here. We may record, however, that they were unarmed, smooth, indehiscent and fusiform, about six inches in length and in shape very like the fruit of *Datura suaveolens*. As is common in some species of tree Daturas, *Methysticodendron* fruits, according to the natives, sporadically every few years but never annually.

The evidence for retaining *Methysticodendron* in the *Solanaceae* seems to me to be overwhelming. Nevertheless, the several fundamental characters in which this concept diverges from the conditions now embraced by the family must be borne in mind. It would seem entirely probable that a new tribe should be set up within the *Solanaceae* to accomodate *Methysticodendron*.

Would we not be warranted in suspecting that we have at hand perhaps a primitive representative of the *Solanaceae*, possibly a prototype of what has to-day developed into the tree-species of *Datura*? The apparent primitiveness in the union of floral parts might conceivably be interpreted in this light. In any consideration that *Methysticodendron* may represent a prototype of the modern representatives of the brugmansioid *Datura*-species, it would be advisable to take into account the interesting fact that the valley of Sibundoy, a locality which we know to be a repository of many curious endemics, is a high Andean region, and the centre of origin of some tree-species of *Datura* is thought to be the northern Andean area.

The Search for a Non-addicting Analgesic

By Harris Isbell, M.D.

From the Addiction Research Center, Public Health Service Hospital, Lexington, Kentucky *

Since potent analgesics are indispensable in clinical practice and at the same time constitute a danger because of their addiction-producing properties, a drug has long been sought which would be non-addicting and at the same time combine the following properties : analgesic potency equal to that of morphine; toxicity no greater and, if possible, less than that of morphine; no undesirable side effects; and at the same time no tolerance to the pain-relieving effect.

Such a substance has been sought since 1929 under the aegis of the Committee on Drug Addiction and Narcotics of the National Research Council. The first approach was to modify the molecule of morphine. This effort shifted to the synthetics with the discovery of pethidine and methadone. In the last few years emphasis has shifted to the analgesic properties of nalorphine and mixtures of opiates and nalorphine. Although nalorphine is itself a modification of morphine, it is an opiate antagonist and has quite different properties. For the time being, the object of this search has not been attained; analgesic potency and addiction liability seem to equal each other.

Six groups of analgesics are known : the morphine, morphinan, pethidine, hexamethyleneimine, methadone, and dithienylbutenylamine families. It is possible, with some imagination, to make out in all six families a central phenylpiperidine nucleus, and it may be that such a structure is necessary for analgesia but is always associated with physical dependence. Such a gloomy view is not necessarily justified. The synthetic analgesics have been discovered, either by preparing a new compound with a structure similar to a drug already known, or by a synthesis made for some purpose other than analgesia. In the latter case, routine testing disclosed that the drug in question induced morphine-like effects in mice and in other animals. Thus new drugs were discovered by their pharmacological resemblance to morphine. Therefore it seems that a new approach could be found : drugs with the phenylpiperidine structure and drugs giving morphine-like effects in lower animals should be avoided and an agent should be looked for which would be analgesic without being otherwise morphine-like; one difficulty in this approach is that it is not easy to deduce the effect on subjective reaction to pain in patients from the analgesic effect in lower animals. It may be that a number of promising drugs have already been discarded because of only weak analgesia in animals; an example of what is meant is nalorphine, which is a very weak analgesic in animal tests but is as effective as morphine in relieving post-operative pain.

Nalorphine does not create physical dependence, but it has undesirable side effects (disturbed mental reactions). Furthermore, it is not known whether tolerance to its analgesic effects would develop with continued use. Investigations are currently under way on possible modifications of the nalorphine structure, which might reduce the side effects while retaining the analgesic effects and the lack of addiction liability.

* The present note summarizes, by permission of the author, a paper he published in the *Journal of the American Medical Association*, 161 : (13) 1254 (July 28) 1956.

Khat

Khat is a shrub the leaves of which are used as a stimulant or a medicine in certain regions of East Africa and Arabia. The question of khat has been placed by the Narcotics Commission on the provisional agenda of its twelfth session, scheduled to meet in April 1957, on the proposal of the representative of Egypt. The following article is a compilation of information from existing published sources on the subject, which are of varying degrees of authority; and it should be regarded as of a background character, pending closer examination of the question by the Commission.

Historical Background

Khat was probably known and used on the Ethiopian uplands, where it seems that it originated, in very ancient times. It is, however, impossible to fix accurately its original habitat and the region where it first developed. As is usual in such cases, some authors state that it was not unknown to classical antiquity. Merab, for instance (58), thinks that it was the smoke of khat that inspired the Delphic pythoness, that Homer's *nepenthe* offered by Helen to Telemachus was none other than khat and that Alexander the Great used it to cure his army of an epidemic disease. There are equally unverifiable suggestions with regard to cannabis and opium, and no importance can be attached to such suppositions.

The first historical reference, as far as could be determined from the literature consulted, occurs in a medieval Arab manuscript (MS.143, Bibliothèque nationale, Paris), where it is stated that the King of Ifat, Sabr Ad-Din decided to plant khat in the town of Marad (the period in question seems to be the first half of the fourteenth century). According to Rochet d'Héricourt (75), khat was introduced from Ethiopia into the Yemen in 1424 by Sheikh Abu Zerbin. Another reference to its cultivation in the fourteenth century in the region of Aden and in the Yemen is found in a sixteenth-century Arab chronicler, Abdul-Kadir. Its cultivation in that region is thought to be earlier than that of coffee (82). Khat was not known to the scientific world until the end of the eighteenth century. During an expedition organized by King Frederick V of Denmark, the physician and botanist Peter Forsskal collected, among many other plants, specimens of khat, which he described under the name of *Catha edulis*. The only survivor of the five members of the expedition, the Hanoverian geographer, Karsten Niebuhr, published the botanical papers in 1775, and in memory of his friend called *Catha edulis* " *Catha edulis forssk* " (65).

In the first half of the nineteenth century, various travellers and scientists visiting Arabia and East and South East Africa referred to and described khat either under that name or variants of it : *Celastrus edulis* Vahl, by Ferret and Galinier (37), *Catha Forsskali A. Richard*, by R. Petit, *Methyscophyllum glaucum*, by Ecklen and Zeyher (27), etc.

Description

Khat is a shrub with persistent leaves of the *Celastraceae* family, relatively of little importance, which includes the spindle-tree, *Celastrus*, etc.; mention may be made, however, of a Philippines plant of the same family, *Lophopetalum toxicum*, containing a poisonous substance lophopetalin, which is used in the composition of arrow-head poisons (67). Khat grows to between 3 ft and 6 ft (1–2 m) in high and dry soils, reaches 20 ft (6 m) on the moist slopes of the Ethiopian mountains and may even in the equatorial region under favourable circumstances reach a height of 80 ft (25 m). The trunk may be some 24 in (60 cm) in circumference and the bark is thin, smooth and brown; in appearance the tree is not unlike the tea shrub.

The new *leaves* are reddish-brown, becoming greenish-yellow when fully grown. They are bifarious, elliptical, lanceolate, sharp, coriaceous and almost tasteless. Dimensions vary widely from 0.2 in (0.5 cm) to almost 3 in (7 cm). The limb, which at the base is smooth, has over the remainder of the leaf short, mossy serrations; the median vein projects on the under, reddish side; the secondary veins meet before the margin of the limb; between the secondary veins the smaller ones form a reticulate venation. From the histological point of view, Perrot (67) gives the following description : " Glabrous epidermis; bi-facial messophyl with two palisade layers and spongy parenchyma with ramous cells of twin crystals, isolated or sometimes grouped in clusters in the same cell. Fasciculate system in an arc almost closed by two superior phlöem-lignous strands. Many periphlöem units. Crystalliginous phlöem ".

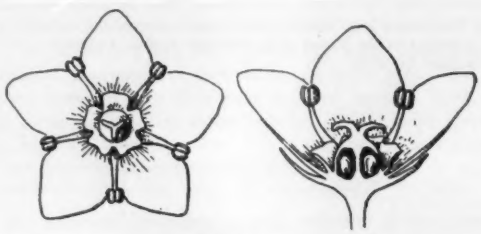


FIGURE 1

Flower of khat

FIGURE 2

Fruit and seed of khat (note the " wing " of the seed)

The *flowers* are small, white or greenish and arranged in axillary cymes. They have five equal sepals, deeply indented, and five oblong petals. The *fruit* is a linear capsule, dark brown, with three valves, each *loculus* containing one to three small *seeds* at the base of a thin brown wing. The seeds, brown-red in colour are about one-eighth of an inch (3 mm) long.

Variants and Vernacular Names

It is generally recognized that the genus *Catha edulis* consists of only one species; however, in view of its extreme polymorphism, it is certain that it comprises several cultural varieties which seem not to have the same properties. A. Chevalier (20) points out that A. Richard noted in Ethiopia a variety which he calls *Celastrus padinus Rich.* with longer petioles and broader limbs. The Natal form, called *Methyscophyllum glaucum* Eckl. and Zeyher is certainly a distinct variety with narrow glaucous leaves; it is called *Catha glauca comb. nov.*

Given this polymorphism and also the many races represented by those who use khat, the plant is known by a wide range of names which designate either the whole shrub or a quality of the product utilized. The Arab name of khat is the commonest—mainly because Arabic is by far the most widely used language of those which refer to the shrub or its parts. This is the term used in Europe, with variants which are simply phonetic transcriptions according to the language (catha, khat, kat, qat, ciat, etc.). Khat is the name designating the whole plant in Somali and in Amharic, (variant : tschat). The name which is, practically speaking, the most important after khat is " mira " or " miraa "—found in Masai, Kikuyu, Tukuyu and frequently in official or scientific texts in South and East Africa.

In view of the confused state of the literature of the subject in this respect, a list of terms used in East Africa to indicate the plant khat is given below (according to Greenway (40)) :

Gazaland

Mutsawhari (Chendao).

Kenya

Kat (Somali); mirungi, muirungi and miraa (Kikuyu); ol meraa (Masai); liss (Mar.); tumayot (Dor.); muraa (Mer.); mairongi (Mt. Kenya); meongi, maonj and miungi (Kamb.).

Nyasaland

Mutsawari (Mlanje); mdimamadzi (Dedza).

Uganda

Musitate (Lumwege).

Tanganyika

Mlonge (Swahili); mulungi (Somali); warfo, warfi and waifo (Mbulu); seri (Fiome); mandoma, mwandama, m'mke and mfeike (Shambaa); mzengo (Nguru); msekera (Kilongo, Uzinza); nangungwe (Mwera); mhulu, muhulu and muhulo (Hehe); liruti (Hehe, Fuagi); msabukinga (Kinga); msuruti, msuvuti and mbungula mabwe (Rungwe); mira and mbungula mabwe (Tukuyu); ikwa (Nyika).

On the other hand, some authors (20, 39, 68, 83) give lists of terms which distinguish the various qualities of khat. The definitions do not always exactly agree and the chief conclusion to be drawn is that in whatever region it is consumed, khat is regarded as are tea and coffee by connoisseurs, the criteria being the part of the plant utilized (leaves, buds, twigs or a mixture of two or all of these forms), the degree of maturity and, consequently, of size (the young shoots and the small red leaves are the most appreciated), and lastly the region of origin. It seems that there are " vintage " khats and, in all probability, the fraud and the snobbery that accompany that idea. Obviously the choice will depend on the use to be made of khat (see below) and also on the price, which varies considerably with the quality of the product.

Chemical and Pharmacological Aspects

The above-mentioned differences are empirical and subjective in character, and no thorough scientific investigation has been made of them because the authors working on khat never had at their disposal the complete collections of samples of leaves, etc., which would allow the establishment of valid conclusions. From the point of view of the effect, it is quite clear that there is little connexion between the small, thin leaf of a bush some 3 ft (1 m) high, growing at 5,000 ft (1,500 m) in an arid soil, and that of a 16-foot (5 m) tree growing in the equatorial zone. In fact, experience in the case of other alkaloid plants shows that the effects vary with the soil and climatic conditions in which the plant grows (and obviously of cultivation, if the plant is cultivated), that

FIGURE 3

Flowering khat twig (from Nyasaland). Note the large size of the leaves

FIGURE 4

Khat twig with fruit (from Cape Province). The leaves are small and elongated

the alkaloid content changes and that under certain conditions some effects may disappear entirely (for instance, the variations in the opium alkaloids). Further practical proof as regards khat is provided by Mustard (62), who reports the introduction of khat into Florida, where it has been successfully cultivated, and where leaves eaten experimentally had no effect on those who chewed them.

It is therefore impossible at this stage to draw up an overall scientific table of the properties of khat. That is why the following approach has been chosen : first, to give a description of what is known of the chemical analysis of the plant and of the effects of its alkaloids, and to deal in a later chapter with the effects in practice upon man of the plant as cultivated.

* * *

The first studies were those of Flückiger & Gerock (86) and of Paul (86) & Mosso (63). The first two isolated an alkaloid that they called cathine, the last an alkaloid to which he gave the name celastrine. In fact, these studies were of little use, for the substances discovered were quite impure. The second stage of research was in 1901 when Beitter (8) claimed to have extracted a certain number of salts of cathine (sulphate, hydrochloride, hydrobromide, salicylate) and described cathine as crystallizing into needle crystals, bitter in taste, with no smell, soluble in ether, alcohol and chloroform. The formula he gave was $C_{10}H_{18}NO_2$. Subsequently, J. Chevalier studied khat and published an important work in 1911 (21). He suggested the possible utilization of khat or cathine in the withdrawal cure from morphine, a suggestion which does not seem to have been followed up.

In 1912 the *Pharmaceutical Journal and Pharmacist* published a communication made to the Pharmaceutical Society by Dr. R. Stockman (79). This important article gives details of his discovery of three alkaloids of khat : cathine, cathidine and cathinine. The author had at his disposal dried leaves, but—because of either experimental obstacles or difficulties inherent in the substances—he obtained only a low and variable yield. In addition to the alkaloids, the leaves contained sugar, tannin, a small quantity of rubber, a wax-like substance, and a small quantity of a yellowish essential oil that was pleasant to smell and to taste. The first alkaloid isolated, cathine—as a sulphate—was in the form of white, needle crystals, of bitter taste, easily soluble in water and diluted alcohol, but not in pure alcohol, chloroform, ether, etc.

Cathidine is a white amorphous powder insoluble in water, soluble in ether, pure alcohol, acetone, etc.

These descriptions were repeated more or less exactly in subsequent publications (up to and including the Merck Index 1940) (59). The proportions given for the three alkaloids (which, as has been seen, must be taken with reserve) are 0.27 per cent for cathine, 0.32 per cent for cathidine and 0.15 per cent for cathinine. Even admitting possibilities of wide error, these percentages are such that commercial extraction does not seem very promising.

In 1930, Wolfes (86) gave for cathine a different formula from that of Beitter (perhaps the latter had isolated the substance in the form of glycoside) : $C_9H_{13}NO_2$. He showed that it is identical with d-nor-iso-ephedrine $C_6H_5CHOH.CH(NH_2)CH_3$. This substance is one of the alkaloids which in the *Ephedra vulgaris* of Japan (Ma-Huang) are the satellites of the *l*-ephedrine used commonly in medicine as a sympatheticocomimetic. Possibly, according to Hesse's theory (46), the other two alkaloids also closely resemble ephedrine.

On account of the difficulties of obtaining leaves of khat, whether fresh or not, and perhaps also because of their small commercial value as shown by the early analysis, original studies on the chemical composition of khat are rare. In 1952, Miss Mustard (62) published the results of research into tropical plants in order to determine their ascorbic acid content. She made five analyses of a mixture of leaves and twigs—" commercial khat "—and found an ascorbic acid content of 135.7 mg/100 g. Analysis of leaves only gave the high content of 324 mg/100 g. Obviously, it might be stated that the khat consumers use it unconsciously on account of the high ascorbic acid content, but even if that were relevant to the consumption of khat, it is probably only one, and not the most important, among many considerations (it is true that scurvy is endemo-sporadic in the region where khat is consumed, but there are in such regions many other plants containing ascorbic acid that are not chewed).

Lastly, Peters (68) draws attention to a very interesting fact : " In a recent series of analyses (1952) carried out by the government laboratories, London, it was demonstrated that there was no appreciable difference between the alkaloidal properties of fresh samples of khat and samples that had been dried and preserved for a period of ten days. It would appear from this that, if indeed there is a difference between the properties of the fresh leaves and those of the dry—and local opinion substantiates this—there is an additional factor to be considered, which might well be a property contained in the resin of the plant."

Just as the habit of coffee drinking cannot be entirely explained by the presence of caffeine, so the chewing of the khat leaves is not exclusively dependent on the alkaloid content. That is why, in order to determine their action, laboratory experiments on the pharmacodynamics of the khat alkaloids are not enough, and why clinical research would have to be carried out on the spot by using the different categories of khat consumed, a long-term task which remains to be undertaken.

For that reason, it seems advisable to describe separately the pharmacodynamics of the alkaloids, as is done briefly below, and the effects of consumption based on empirical observations of those who have seen khat users. These effects will, therefore, be described in the following chapter after a summary of the methods of cultivation and consumption.

* * *

One of the first to carry out accurate experiments on khat, Leloup (49) about 1890, found that the substance excited the nervous system, delayed sleep and increased muscular strength. His experiments, carried out on himself, are extremely subjective.

In 1911, J. Chevalier (21) experimented on animals with cathine that he had isolated; according to him, small doses induce excitation of voluntary movements but not reflex excitability. He gave 3 mg as a " weak dose " and 6–10 mg as " lethal doses ". (Unfortunately the animals dose/weight ratio is not given, and the author refers merely to a " large " frog.) In the initial stage of excitability, the animal jumped about, foaming at the mouth, and then calmed down, showing symptoms of paralysis; sensitivity was retained, but motor action and reflex excitability gradually disappeared. The heart slowed down and then stopped in systole. These phenomena were more marked with guinea-pigs and rabbits, and, after an intravenous injection of 3–4 cg/kg, death took place in three or four hours, accompanied by convulsions and muscular rigidity. Smaller doses produced signs of excitation, with dilation of the pupil, rapid breathing, etc. Dogs are more resistant, and 5–6 cg/kg are required to bring on convulsions.

Stockmann also conducted experiments with these alkaloids, but his account (79) does not give any quantitative determinations. He states that cathine has an action on the nervous and muscular system of the frog similar to that of a combination of morphine and caffeine. Cathinine is a stimulant, rather, and cathidine is both a " muscular poison " and a stimulant. It is interesting to note that an infusion of khat leaves in water produces the same effects as cathine, cathidine being insoluble in water and acting only if the leaf is chewed, and not drunk as an infusion.

As far as is known to the writer of the present paper, the pharmacopoeias consulted do not appear to mention khat or its alkaloids except that the British Pharmaceutical Index of 1949 refers to khat (p. 902) as one of the " substitutes and adulterants of tea ". However, the following commercial experiments were undertaken : in 1910, a Lyons pharmacist introduced a speciality based on khat, which he called the " New Abyssinian tonic " which, as its name indicates, was sold as a restorative, but he was unable to obtain sufficient supplies of raw material and production came to an end at the outbreak of the first World War (76); in 1913, a London pharmacist began to manufacture products based on khat. Dr. Martindale (2) marketed three products : " (1) catha-cocoa milk, (2) catha-cocoa glycero-phosphate... a nervous tonic and stimulant in which milk powder is combined with catha extract and calcium glycero-phosphate, and (3) effervescent phenol-phtalein with catha... a mild tonic laxative. Extract of catha is also put up in tablet form. "

Dr. Martindale said that he procured khat leaves in Arabia, but that recently (1936) he had had some difficulty in obtaining a supply. The demand for the product was " general, but limited ". He exported a certain amount of his products to India. During more than ten years' experience of selling his preparations, he had never had any complaints or heard of any bad effects. Neither the 1948 edition of the British Pharmacopoeia nor the British Pharmaceutical Index of 1949 mentions these preparations.

Habitat and Cultivation

Khat is found wild in the relatively humid mountainous regions of East and South Africa, but not, it seems, in Southern Ethiopia, although it is cultivated there; in natural conditions it grows at a height of from 5,000 to about 6,500 ft (1,500–2,000 m). As a rule, it grows in association with conifers. It is found in the following regions : *Arabia :* North Hedjaz and East Hadramaut (not confirmed) (20); *Belgian Congo :* Sake region—near Lake Kivu— Albert Park, Ruwenzori region, Lake Mujunga; *Eritrea :* information is contradictory, some authors stating that it is found here, while others deny it; *Ethiopia :* regions of Abba Yerma, Axum, Adowa, Chiré, Choa; *Kenya :* Northern Frontier District from Mount Kenya to the Chyulu Hills (17); *Nyasaland :* Dedza district, Mount Mlanji, Blantyre district (31); *Uganda :* Kigezi, Karamoga, Bugishu districts, region of Mounts Oebasian and Elga (29); *Southern Rhodesia :* regions of Salisbury and Umtali (34); *Tanganyika :* Khat has been reported in practically all mountain districts at heights of from 4,000 to about 8,000 feet (1,200–2,500 m) (4); *Union of South Africa :* region of Queenstown.

It may be assumed that in most of the above-mentioned regions growth is sporadic. However, in addition, there are references to other regions that do not specify whether the plant is cultivated or grows wild (e.g., Afghanistan, Turkistan, etc.). The main zones of genuine cultivation are Ethiopia (particularly Harar), the Yemen, the region of Aden and Hadramaut and Kenya. Attempts at cultivation have been made in many parts of the world, either in the open (Ceylon, Bombay district, Florida, Algeria, Portugal, France (Villa Thuret at Cap d'Antibe)), or under glass in the various botanical gardens. Obviously khat grows best where the conditions are most like those of its normal habitat—i.e, on fairly moist slopes between 5,000 and about 8,500 ft (1,500–2,500 m), depending on the latitude.

The literature on the subject is full of contradictions with regard to methods of planting, maintenance and harvesting, which may be due—particularly when found in travellers' writings—to too hastily gathered information or to the impossibility of collecting details, such as questions of reproduction by seeds or cuttings, number of crops, etc. The description given by Peters (68) and cited below is one of the most recent and, besides, covers practically all the points of earlier observers, and the author states that in order to write his articles he visited the Harar plantations, which are probably the largest. It may be taken that, *mutadis mutandis*, his description will fit all plantations :

" Khat grows best at an altitude of between 5,500 and 8,500 ft (1,650–2,500 m); the Harar plantations are in the region of 6,000 ft (1,800 m). The trees are to be found growing in rows of five or six, with rows of coffee trees interspersed with the khat trees; millet was found growing in the spaces between the trees. Artificial irrigation is not employed, the trees relying entirely on rainfall. They are cultivated from cuttings; seed is sometimes, though rarely, sown. The cuttings are planted and artificially watered for a period of approximately 40 days : they are then allowed to grow to three or four years old before the first crop is taken; they then yield two crops a year. The average height of the trees is about 16 ft (5 m), and the older trees, from which the main crops are obtained, are about twenty to twenty-five years old.

" The trees are pruned yearly to keep their height to approximately 16 ft, as without this they have been known to grow to well over 60 ft (20 m). No disease seems to be known amongst the khat trees on the plantations visited, and although adjacent coffee trees have been known to die, no disease is transmitted to the khat trees. The trees are grown and tended with the same primitive agricultural implements as used by the farmers' ancestors for many centuries. The khat is picked two or three times a week and sent to the markets in Harar, where it is accepted as fresh for a period of up to four days after picking. After this, it depreciates in value, and, apparently, in potency."

The usual method of transporting khat for sale is to make it up into small packages, the most common measure being the bundle of 40 twigs of approximately 16 in (40 cm) long, wrapped in leaves and carried to the market either by caravan or, nowadays, by rail or air. There are few details known to the present writer of the extent of the trade and the price paid. The following figures were available : Tanret (80) mentions a price of 20 French francs a kilogramme (1933) in Ethiopia and it was said that in certain villages of the region of Aden the men consume daily two U.S. dollars' worth of khat.[1]

* * *

It is difficult to evaluate the fluctuations of khat cultivation. It is, however, interesting to note that a document submitted to the sixth session of the Eastern Mediterranean Regional Bureau of the World Health Organization (document EM/RC6/5, 9 July 1956) stated that its use was spreading so rapidly that cultivators grew khat instead of food and other cash crops.

Thus the cultivation of khat has become more and more important, and the governments of the different areas in which it is grown have taken measures to control it, principally by way of taxation and registration of the users. The first attempt seems to have been made in British Somaliland (4), where in 1921 the cultivation of khat was prohibited, as was its sale, except by licensed vendors, who were permitted to sell it only to properly registered consumers. Owing to difficulties of application, however, that measure was subsequently withdrawn. In 1939 there were further measures of control in British Somaliland providing for right of search and penalties of imprisonment and fine for its cultivation.

In certain districts of Kenya the possession of khat was made an offence (3) : the council of the Isiolo district took that decision in 1934, and in 1935 the native council of the Maru district adopted a resolution prohibiting the use of khat by those who were not inveterate consumers. There is practical control at the present time in Kenya through a system of taxation similar to that on alcohol. The annual report of the Kenya Department of Agriculture (3) for 1947 states that after khat (miraa) had been controlled a quantity to the value of £92,793 was exported from the Maru district of the Central Province. From the point of view of the district revenue, khat was the second most important export item (this sum apparently refers to the period beginning April to the end of the year, 1947). Several authors state without giving details that the tax on khat constitutes an important source of revenue for the Government of Yemen.

It may be added that, although there is no regular trade or exploitation, khat wood is used for the construction of dwelling huts. Moreover, in view of its golden-brown colour, its very fine texture and the fact that it is easily polished, it has been suggested (29) that it might be used for joinery and marquetry. According to Greenway it gives a pulp out of which excellent blotting paper could be made (40).

Many of the authors have suggested that apart from the taxation measures there should be measures of control in view of the dangers presented by the consumption of khat. Such a suggestion is found, for instance, in the *Pharmaceutical Journal* of July 1952.[2]

There have been indications in the French press that measures of control were being taken by the French Government.[3]

Khat is not covered by the conventions relating to narcotic drugs. The Advisory Committee (of the League of Nations) on Traffic in Opium and Other Dangerous Drugs, however, took up the question at its twentieth session. In considering the survey of the annual reports of governments, the Chairman of the Committee referred to the mention of khat in the annual report of British Somaliland and asked the United Kingdom representative to supply further information on the matter. On 16 November 1935, the United Kingdom representative communicated to the Secretariat two reports on the question : one from Dr. Donaldson, Medical Senior Officer, British Somaliland, the other from Mr. A. W. Exell, of the Natural History Department of the British Museum (2).

The question was not taken up again internationally until the eleventh session of the Commission on Narcotic Drugs (23 April - 18 May 1956). The question was placed on the provisional agenda of that session at the request of the representative of Egypt who communicated a report (1) from the laboratory of the Egyptian Medico-legal Institute. This document gives the general characteristics of khat and provides some information on its use in the Yemen. The Commission decided to defer the study of the question of khat until its next session and placed it on the provisional agenda for that session (item 11)[4] expressing its intention of inviting Ethiopia and Yemen to participate in the discussion.

---

[1] *Long Island Daily Press*, 16 August 1954.

[2] *Pharmaceutical Journal*, Vol. 169, 4629, page 55.

[3] See *Le Monde*, 7 December 1956, page 5.

[4] Commission on Narcotic Drugs : *Report to the Economic and Social Council on the Eleventh Session of the Commission*, document E/2891 E/CN.7/315.

Methods of consuming Khat

Methods of consuming khat vary according to a certain number of factors : the regions concerned, and of course, the kind of khat that the consumer has available, the price he can pay and, lastly, the purpose of using it—that is to say, his own personal tastes. Fifty years ago the methods were relatively the same for all the regions, since transport, being by caravan, was slow, and, consequently, khat was necessarily consumed dry by those who did not cultivate it. The methods have changed since transport is now either by rail or, particularly, by air. Whereas nineteenth-century travellers and explorers nearly always described the Arabian use as being in the form of infusions of the dried leaf, nowadays this method seems to be giving way to mastication of the fresh leaf, since khat is imported to Aden and the Yemen and elsewhere by air from Harar. This shows that the methods of consuming khat were not necessarily the result of choice or particular taste, but depended rather on circumstances.

The three normal methods of consuming khat are : (1) *chewing*—in this case the fresh plant is used—i.e., khat picked four or five days previously (at most), which has kept its moisture, since for that purpose it has been wrapped in leaves which are moistened from time to time. The more tender and juicy the leaves (either because they are taken from a plant variety with small leaves or because they are picked from the end of a branch), the easier it is to chew them and consequently the more they are appreciated. Thus, according to Peters (66), there is in Amharic a special name for this kind of leaf (koda) and also in Somali (hagafa). According to Vaughan (83), this kind of khat, called malhani, is more sought after in Southern Arabia, Aden, the Yemen, Hadramaut, etc. According to the size and the freshness of the leaves, therefore, there are " vintages " of khat, which are highly appreciated by the connoisseurs, and the price of which varies with the demand. A consumer used to good-quality khat chewed fresh would find it as difficult to get used to the dried leaves as, for instance, a cigar-smoker would to chewing tobacco. When the khat to which the consumer is accustomed is unobtainable, he would rather go without than try an inferior quality.

To chew the khat, the twigs are stripped of their leaves, which are made into a kind of quid, which is chewed until all the juice is extracted. To help the process, the chewer drinks large quantities of water. The process may last ten minutes or a quarter of an hour; when it is over, the residue is swallowed. During chewing, cigarettes are sometimes smoked. It may be remarked that when the leaf is fresh, the saliva is sufficient to extract the active elements—the alkaloids alone or combined with some other part—whereas with the dried leaves chewing alone does not seem to be sufficient.

The second method consists in using the large leaves which would be too hard for chewing or the leaves that have lost their freshness on too long a journey. In this case, the leaves are used dry—either the large leaves after drying in the sun or the others used as received from the seller. The dried leaves are powdered in a mortar; the powder is placed in a bowl and, after adding sugar and sometimes spices and water, a kind of paste is made, which is then chewed. As before, the residue is swallowed—and large quantities of water are drunk—probably to facilitate the extraction of the active elements. It may be added that, according to individual taste or to regions—always where fresh leaves are not available—a khat infusion is made in the same way as tea. This method is not very common and is followed mainly in South Africa (64). It is more usual to add to ordinary tea fragments of khat twigs to alter the taste and the effect of the tea. It may be pointed out that with infusion, at least one of the alkaloids, cathidine, is lost, being insoluble in water, for so far as is known the residue of the infusion is not consumed. Since it seems that the effect is not different from that produced by chewing, it may be concluded that cathidine is not responsible for the effect produced by khat, although the action of khat is still not very well known. Merab (58) points out that the Ethiopian Christians use khat in preparing certain of their beverages (tedgee and talla).

Lastly, in certain parts of Arabia, khat is smoked like tobacco or cannabis, the ends of the twigs and the leaves being first crushed and then rolled in cigarettes (no mention of khat smoked in a narghile has been found).

The taste for the different varieties of khat seems as strange to those who are not accustomed to it as the choice between different qualities of wine or tea to non-initiates : it has probably not much to do with the alkaloid content. Khat is taken not only for its stimulating or inebriating effect, but also for the pleasure given at the time of taking it. From that point of view, therefore, it must be ranked with substances such as tea, tobacco, alcohol, etc., at any rate as regards occasional use. According to observations made on the spot it appears that after some time, once the need has been created, khat is no longer used for the pleasure derived from its taste, but mainly or solely for its effect; that marks the transition between the occasional and the habitual user. It may be observed that it is about the same with alcohol, which is taken by the occasional consumer for the pleasure derived from it, whereas the confirmed drunkard seeks principally or solely for the effect, whatever the taste or the appearance of the product consumed. In this connexion, some authors have wondered whether the real difference between occasional consumption, which might be styled pleasure or " sociability " consumption, and harmful habits does not lie precisely in the fact that the pleasure, which is sought at the time of consumption, is complex in nature—taste plus knowledge of the effect—whereas in the case of a harmful habit there is mainly or exclusively the craving for the effect.

Before considering the effects of khat, it may be remarked that it is consumed—when it is consumed for pleasure—in a group; that is to say it is a part of social life. In almost all the regions of habitual use, travellers mention this social habit—for instance, the fact of offering the stranger or the guest a twig of khat to chew or to welcome him in offering him a cup of khat, just as elsewhere he would be given a cup of coffee or a cocktail. Moreover, khat is very frequently taken socially, either in shops or public places where men meet to pass the time of day, or in private houses, where one is invited to " take khat ". Here again, as with alcohol and the like, there is a marked difference between the occasional consumer, who uses it in company, and the inveterate consumer who looks for the effect, and for whom in consequence the social aspect is a drawback rather than an advantage.

Effects of Khat

The foregoing analyses show that the effects of khat are quite different on the occasional and on the habitual consumer. On the former they will be strictly proportionate to the quantity of khat taken. The case of the latter is much more

complex. First, the effects obviously include the consequences of the khat taken as such—and the habitual user consumes increasingly large quantities in order to obtain the same effect; then there is a whole series of secondary *physiological* effects. The inveterate consumer loses his appetite and eats less and less; his body becomes emaciated, and he falls a victim to various deficiency diseases, not to mention secondary complications, such as constipation, etc. Among the *psychological* effects, the person who sleeps and eats less falls a victim to various illnesses. He clearly loses intellectual and will power. That situation gives rise to certain *social consequences*. The inveterate consumer no longer bothers about himself, his family or the rest of society. If he has the means for procuring khat, he becomes a mere human wreck. If he has not got the necessary money, he turns to begging or some kind of anti-social activity, such as crime.

Little is known of the effect of khat on a normal individual without considering the period of use. Since it has not been thoroughly studied from the scientific point of view, but only empirically observed by travellers or, less frequently, reported by those who have taken it occasionally, the descriptions of it are sometimes contradictory. Generally speaking, there is a kind of exaltation, beginning with a feeling of being liberated from space and time, accompanied by extreme loquacity and inane laughter, until a semi-coma intervenes, out of which the consumer awakes in a painful condition. In the first stage, therefore, there is intellectual excitation with euphoria, in the second stage the numbing of the intellectual faculties, and in the third the eclipse of these faculties —lack of attention and memory accompanied by the liberation of violent instincts.

A distinction must be made between occasional and chronic use :

In the case of *occasional use* khat abolishes sensations of fatigue and hunger, soothes passions such as anger, then degenerates into a state of veritable poisoning, accompanied by dilatation of the pupil, cold sweats, convulsions, hyperaesthesia, etc. and, if taken in excessive quantities, may even result in death. (Heisch (44) describes a specific case that he had observed of a khat-chewer who, when taken to hospital, showed the above-mentioned symptoms, then passed into a coma and died after four days. At the post-mortem, the stomach was discovered to be full of khat and death could not be attributed to any other cause than its consumption.)

In the case of *chronic use*, khat numbs the intellectual faculties and leads to complete stupor or madness. Carrothers (17) reports two cases of madness brought on by khat in which he observed incoherence, feverish excitation, marked troubles with a schizophrenic character and, finally, manifestations akin to *delirium tremens*. Incidentally, both patients quickly returned to normal when the khat was withdrawn.

To sum it, there is a quasi-medical use of khat : it has been used for a long time by couriers carrying urgent messages as sustenance during the journey when there was no time to stop for eating or sleeping. It is also used in traditional medicine in Ethiopia, Arabia and elsewhere for all sorts of purposes—e.g., as a diuretic in the treatment of gonorrhoea—as a prophylactic against malaria, asthma, coughing and diseases of the chest, stomach trouble and even as a preventive against plague. This list shows that in the therapeutic practice of primitive traditional medicine khat becomes a panacea.

Khat has also a *religious or magic* significance. According to Tanret, (80) Mgr. Jarosseau, Bishop of Harar, noted this phenomenon in the following terms : " The whole country is persuaded that this shrub enjoys divine blessing; no private or public religious ceremony can take place without the ritual chewing of the leaf accompanied by much praying and singing. In the Moslems of Harar it induces a strong religious exaltation, which they regard as a gift from heaven."

There is again the use of khat for pleasure. Since khat is regarded as an excitant and a stimulant like coffee, tea and the like, it is generally not considered prohibited by Koranic law and consequently there is no moral objection in the Moslem countries affected against its abuse.

Khat is used in relatively poor regions, where the frequent lack of food encourages deficiency diseases. Consequently, in view of its stimulating effect, it is often not unnaturally regarded as a divine blessing.

For populations with relatively few sources of distraction it constitutes a simple and convenient form of escapism.

The observations, however superficial, that have been made show that khat may produce a definite intellectual and moral deterioration and that its social consequences may be serious.

Except in sporadic cases such as the utilization of the wood of the shrub, it is generally not used for industrial purposes.

Bibliography of khat

1. UNITED NATIONS COMMISSION ON NARCOTIC DRUGS. *The problem of khat*, Document E/CN.7/L.123, 20 April 1956.
2. LEAGUE OF NATIONS ADVISORY COMMITTEE ON TRAFFIC IN OPIUM AND OTHER DANGEROUS DRUGS. *Reports on the characteristics and uses of the plant kat (Catha edulis)*. Document O.C.1617, 3 February 1936.
3. Annual Report of the Kenya Department of Agriculture, 1947, p. 45.
4. Anonymous. The need for the control of khat. *East African Medical Journal*, 1945, *22* : 1, pp. 9–10.
5. Bailey, L. H.; Bailey, Ethel Zoe. *The Standard Cyclopedia of Horticulture*, Macmillan, New York, 1947.
6. Baird, D. A. *British Medical Journal*, 1951, pp. 15–22.
7. Bally, P. R. O. Catha edulis, *East African Medical Journal*, 1945, *22*, pp. 2–3.
8. Beitter. *Archiv der Pharmazie*, 1901, *239*, p. 17.
9. Bertheraud. *ournal de médecine et de pharmacie de l'Algérie*, 1889, p. 151.
10. von Bibra. *Die narkotischen Genussmittel*, 1855.
11. Blatter, E. Flora Arabica. *Records of the Botanical Survey of India*, 1923, *8* : 4, pp. 459–461.
12. Bois, D. *Les plantes alimentaires chez tous les peuples à travers les âges*, Lechevalier, 1937.
13. Botta. *Relation d'un voyage dans l'Yémen (*1837*)*, Paris, 1841.
14. *British Pharmaceutical Codex*, 1934, p. 1048.
15. Britton, Everard, B. A Journey to the Yemen; the Use of Qat. *Geographical Journal*, 1939, *93*, pp. 121–122.
16. Bruecke, F. Th. Ueber die zentralerregende Wirkung des Alkaloids Cathin (The central stimulating effect of cathine), *Archiv für experimentelle Pathologie und Pharmakologie*, 1941, *198*, pp. 100–106 (1 fig.).
17. Carrothers, J. C. Miraa as a Cause of Insanity. *East African Medical Journal*, 1941, *22*, pp. 4–6.

18. CASTELLANI, E. Prime natozie sulla rogna del Ciat (First notes on a canker of khat), *Nuovo Giornale bot. Ital.*, 1946, *53* (1/2), pp. 346–349 (1 fig.).
19. CERULLI, E. *La Lingua e la Storia di Harar*, Pubblicazioni dell' Istituto per l'Oriente, Rome, 1936.
20. CHEVALIER, A. Les qat's d'Arabie, d'Abyssinie et d'Afrique orientale, *Revue int. Bot. appl. Agron. trop.*, 1949, *29* : 322–323, pp. 413–417.
21. CHEVALIER, J. Etude pharmacodynamique du *Catha edulis*, *Bulletin de la société pharmacologique*, 1911, XVII, p. 264.
22. CHIOVENDA, E. *Raccolte botaniche fatte dal missionari della consolata nel Kenya*, p. 21, 1935.
23. CHRIST. *Archiv der Pharmazie*, 1870, *141*, 67.
24. COLLIN. *Journal de Pharmacie*, 1893, sér. V, vol. XXVIII.
25. DEFLERS. *Voyage en Abyssinie*, Paris, 1889.
26. DEZANI. Ricerche farmacognostiche sulla *Catha edulis*, *Arch. di Chimica, Farmacognosia e Scienze Affini*, 1918.
27. ECKLAR, ZEYHER. *Enumeratio plantarum Africae australis extratropicae*, 1834.
28. EGASSE. *Bulletin de Thérapeutique*, 1889, vol. *117* : 168.
29. EGGELING, W. J. *The Indigenous Trees of Uganda*, p. 91, 1941.
30. *Encyclopaedia Britannica*, 11th edition, Vol. 2, 261, 264 (1910).
31. ENGLER, A. *Die Natürliche Pflanzenfamilien*, Vol. 20*b*, pp. 154–155, Wilh. Engelmann, Leipzig, 1942.
32. ERDTMANN, G. *Pollen Morphology and Plant Taxonomy*. Angiosperms (An introduction to palynology, I), p. 104. Almquist & Wiksen, Stockholm, 1952.
33. *Extra Pharmacopoeia*, 23 ed., Vol. 1, p. 295, 1952.
34. EYLES, F. A Record of the Plants collected in Southern Rhodesia. *Trans. R. Society S. Afr.*, 1915, *5* : 404.
35. FAIRCHILD, David. *Exploring for Plants*. Macmillan, New York, 1930.
36. *The World was My Garden*, Scribner's Sons, New York, 1943.
37. FERRET; GALINIER. *Voyage en Abyssinie*. Atlas Botanique, Paris, 1847.
38. GLASER. Travel in Arabia. *The Geographical Magazine*, 1887, p. 291.
39. GLOVER, P. E. *A Provisional Check List of British and Italian Somaliland Trees, Shrubs and Herbs*. MS. 1946.
40. GREENWAY, P. J. Khat, *East African Agricultural Journal*, 1947, *13*, pp. 98–102.
41. HAKIM, S. *British Medical Journal*, 1951, p. 852, p. 1522; Indian Remedies for Poor Memory, *ibid.*, 1952, p. 222.
42. HARRIS, C. *The Highlands of Ethiopia*, II, p. 414, London, 1844.
43. HARVEY, W. H.; SONDER, O. W. *Flora Capensis* I, pp. 463–464, 1860.
44. HEISCH, R. B. A Case of Poisoning with *Catha edulis*, *East African Medical Journal*, 1945, *22*, pp. 7–9.
45. HENRY, T. A. *The Plant Alkaloids*, 1924.
46. HESSE, E. *Narcotics and Drug Addiction*. Philosophical Library, New York, 1946.
47. LAMARE. *La Géographie*, pp. 34–64, Paris 1931.
48. LEFÈVRE, PETIT, QUARTIN-DILLON. *Voyage en Abyssinie, 1839–43*, Vol. IV, p. 134.
49. LELOUP. Le *Catha edulis*, *Bulletin de Thérapeutique*, 1890, *119*, p. 216.
50. LEWIN. *Phantastica*, Berlin, 1927.
51. LINDLEY, J.; MOORE, T. *The Treasury of Botany*, I, pp. 239–310.
52. LINDLEY. *Vegetable Kingdom*, London, 1846.
53. LOSENER. *Notizbl. Bot. Gart. UMS*, Berlin-Dahlem, 1939, XXIV, p. 450.
54. LORENZO VELASQUEZ, B. Benzedrine, Methedrine and Cathine, *Farmacoterapia actual* (Madrid), 1944, *1*, pp. 5–11.
55. LUM, E. A. *The Manufacturing Chemist*, 1948, *19*, p. 98.
56. MARTINDALE. *Extra Pharmacopoeia*, I, p. 325, 1914.
57. MASCRE, M. Matières odorantes et matières médicinales de la France d'outremer, *Produits pharmac.*, 1947, *2* : 9, pp. 392–403.
58. MERAB. *Impressions d'Ethiopie*, Leroux, Paris, 1921–1929.
59. *Merck Index*, 5th ed., p. 126, 1940.
60. METCALFE, C. R.; CHALK, L. *Anatomy of the Dicotyledones*, Vol. I, pp. 387–397, Clarendon Press, Oxford, 1950.
61. MORI, M. Notizie sul ciat ethiopico, *Agric. coloniale* (Florence), 1940, pp. 201–203.
62. MOSER, C. The Flower of Paradise, *Nat. geogr. mag.*, 1917, *173*.
63. MOSSO. Azione Fisiologica del principio attive de *Cel. edulis*, *Rivista Clinica*, 1891, 30, p. 65.
64. MUSTARD, Margaret J. Ascorbic Acid Content of some Miscellaneous Tropical and Subtropical Plants and Plant products. *Food Research*, 1952, *17*, pp. 31–35.
65. NIEBUHR, C. *Flora aegyptisco-arabica*, p. 75, 1775.
66. OLIVER, D. *Flora of Tropical Africa*, I, pp. 364–365, 1868.
67. PERROT, E. *Matières premières du règne végétal*, II, pp. 1331–1334, Masson et Cie, Paris, 1943–1944.
68. PETERS, D. W. A. *Pharmaceutical Journal*, 1952, *169*, pp. 17, 36.
69. PETRIE, P. W. R. *Journal of Tropical Medicine and Hygiene*, 1939, *42*, p. 357.
70. PLANCHON; COLLIN. *Les drogues simples d'origine végétale*, II, p. 596, Paris, 1896.
71. PLOUVIER, V. Nouvelles recherches sur le québrachitol des Sapindacées et Hippocastanacées, le dulcitol des Célastracées et le saccharose de quelques autres familles, *C. R. Acad. Sci.* (Paris), 1949, *228* : 24, pp. 1886–1888.
72. POIROT. *Hortus Thuretianus*, Paris, 1913.
73. REITTER. *Pharmakognostisch-chemische Untersuchung der Catha edulis*, Strasbourg, 1900.
74. RENDLE, A. B.; BAKER, E. G. *et al.* A Contribution to our Knowledge of the Flora of Gazaland, *Journal Linn. Soc. Bot.*, 1911, *40*, p. 44.
75. ROCHET D'HERICOURT. *Voyage en Abyssinie*, Paris, 1895.
76. SANTAGATA, F. *L'Harar*, Garzanti, Milan, 1940.
77. SCHORLEMMER. *Chemical News*, 9 Nov. 1883.
78. SKINNER, Robert P. *Monthly Consular and Trade Report*, Washington D.C., 1909.
79. STOCKMAN. *The Pharmaceutical Journal and Pharmacist*, Nov. 1912, *89* : 2563, p. 676.
80. TANRET. Le Katt, *Presse médicale*, 1933, *22*.
81. TSCHIRCH, A. *Handbuch der Pharmacognosie*, 1917–1926.
82. UKERS, W. H. *All about Coffee*, 2nd Ed., pp. 12, 661, 662, 1935.
83. VAUGHAN, J. *Notes upon the Drugs observed at Aden*, quoted in R. F. BURTON's *First Footsteps in East Africa*, pp. 67–69, Everyman's Library, 1st ed., 1910, reprinted 1924.
84. WATT, J. M.; BREYER-BRANDWIJK, M. G. *The Medicinal and Poisonous Plants of Southern Africa*, pp. 110–111, London, Livingstone, 1932.
85. WEHMER, C. *Die Pflanzenstoffe*, pp. 455–805, Verlag Fischer, Iéna, 1911.
86. WOLFES, O. *Archiv der Pharmazie*, 1930, *268*, pp. 81–83.

Experimental Evaluation of Analgesia

1. New algesimetric method
2. Comparative study of the analgesic action of morphine, pethidine, l-methadone and levorphanol

By C. Radouco-Thomas, M.D.,
Lecturer, University of Geneva, Faculty of Medicine
S. Radouco-Thomas, Ph.D., and Gl. Nosal

To W. Koll and K. Soehring, for their work on the experimental evaluation of analgesia.

CONTENTS

	Page
INTRODUCTION	14
METHODS	15
I. *Experimental conditions*	15
1. Painful stimulus	15
2. Pain receptors	15
3. Substances studied	16
II. *Apparatus*	16
1. Stimulating circuit	16
(A) Generator	16
(B) Electrodes	16
2. Accessories	17
III. *Experimental procedure*	17
1. Preliminary phase	17
2. Determination of the Pain-Response Threshold	17
3. Evaluation of analgesic potency by the increase of the pain-response threshold	18
IV. *Biometric analysis of results*	18
RESULTS	18
I. *Biological response*	18
1. Qualitative study	18
Somatic physiological reactions : R_0, R_1, R_2	19
2. Quantitative study of the Pain-Reaction Test (P.R.T. = R_4)	20
A. Threshold voltage and biological factors	20
(*a*) Inter-animal variations	20
(*b*) Intra-animal variations	21
B. Threshold voltage and biophysical factors (parameters of the current)	22
(*a*) Threshold voltage/impulse frequency ratio	22
(*b*) Voltage threshold/impulse duration ratio	22
II. *Evaluation of analgesic action*	22
1. Narcotic analgesics (morphine, pethidin, l-methadone, levorphanol)	22
A. Qualitative study	22
B. Quantitative study	22
(*a*) Dose/effect/time ratio	22
(*b*) Dose/effect ratio	25
Margin of safety	25
2. Other depressants of the central nervous system (ether, thiopental)	25
DISCUSSION	26
I. *Reliability of the method*	26
1. Physiological data	26
A. Are the reactions evidence of pain?	27
B. Qualitative and quantitative stability of the P.R.T.	27
2. Pharmacological data	28
A. Reliability of the method	28
B. Selectivity of the method	29
Intensity of analgesia	29
Duration of analgesia	29
C. Electivity of the method	30
II. *Validity of the method*	30
1. Comparison with results of animal experimentation	30
A. Electro-dental methods	30
B. Thermo-cutaneous methods	32
2. Comparison with results obtained in man	33
A. Experimental algesimetry (superficial, deep and visceral pain)	33
B. Pathological pain	34
III. *Efficiency of the method*	34
CONCLUSIONS	34
ACKNOWLEDGEMENTS	35
BIBLIOGRAPHY	35

Introduction

Doch, einzig in der engen Höhle
des Bakenzahnes verweilt die Seele.
Wilhelm Busch

The " pain complex " comprises a primary, sensory process—the sensation of pain—with, superadded, a series of " psycho-physiological " alarm and defence reactions.

Pain is not a normal condition and it is actually injurious when chronic. " Pain itself may initiate or perpetuate a biological destructive cycle ", Wolff 1951 (175). Man has always sought to suppress it.

Among the many monographs and works on the physiopathology of pain and analgesic medication, mention may be made of the following : Kruger, Eddy & Sumwalt, 1940 (95); Lewis, 1942 (103); Symposium on Pain, 1943 (152); Wolff and Hardy, 1947 (174); Symposium on New Synthetic Analgesics, 1948 (153); Isbell, Fraser and al. (76, 77, 78, 79); Wikler, 1950 (161); Wolff & Wolf, 1951 (175); Beckett,

1952 (9); Dorpat, 1952 (35); Schaumann, 1952 (133); Bonica, 1953 (15); Hardy, Wolff & Goodell, 1953 (56); Schoen, 1953 (137); Wikler, 1953 (162, 163); Braenden, Eddy & Halbach, 1954 (18); Jacob, 1954 (82); Scherrer et al, 1954, 1955 (134, 135); White & Sweet 1955 (160 b); Radouco-Thomas C., Radouco-Thomas S. & Nosal, 1956 (123, 124, 124b). There are also recent works by Hazard (60), Jacob (83) and Scherrer (136), published under the direction of Alajouanine, 1957 (1), on pain and its pharmacodynamics.

A survey of antalgic medication as now practised shows that it is fundamentally symptomatic. Two groups have pride of place in the arsenal of pain therapeutics—the non-narcotic analgesics (antipyretics) and the narcotic analgesics (morphine and the morphinomimetics).

The first group has a moderate and partial antalgic action, often attended by toxic side-effects. The second comprises highly potent analgesics as effective as they possess addiction liability.

It is to pharmacology and experimental therapeutics that falls the difficult task of selecting by appropriate methods ever more potent and more easily tolerated drugs from the amorphous mass of new synthetics.

The much-debated problem (44, 82, 146, 160b) of the value of the algesimetric methods used in experiments with animals and on man will be discussed later on. Suffice it to state here that there are now more and more neuro-physiological and neuro-pharmacological arguments for the view that the real evidence for the sensation of pain in animals lies in a " chain " of physiological reactions (somatic-vegetative-humoral).

Many different methods are used for the experimental evaluation of analgesic action.

Chemical and physical stimuli (mechanical, thermal and electrical) have been applied at the level of the various pain receptors to reproduce the classic types of pathological pain (superficial, deep and visceral) in animals. A number of studies have been devoted to this problem by Seevers et al., 1936 (140), 1937 (141), Goetzl, Burril & Ivy, 1943 (50); Pfeiffer et al., 1948 (120); Miller, 1948 (114); Wirth, 1952 (170); Fleisch & Dolivo, 1953 (41); and Jacob, 1954 (82).

Among the multitude of algesimetric methods, our attention has been particularly drawn to the electro-dental stimulation introduced into experiments with animals (dogs) by Koll & Reffert in 1938 (91), and later used and improved by Goetzl et al., 1943 (50), Soehring et al., 1949 (144) and Boréus & Sandberg, 1955 (17). Though it has given remarkable results, it remains confined to specialized research : " Die Methode von Koll hat den besonderen Vorteil am höher organisierten Tier zu arbeiten. Sie erfordert aber bezüglich Tierhaltung, operativer Vorbereitung, u.dgl. einen erheblichen Aufwand, so dass sie mehr für spezielle als für Reihenuntersuchungen in Frage kommt."—Wirth, 1952 (170).

We venture to propose a new routine algesimetric method based on electro-dental stimulation of the pain-receptors of guinea-pigs.

This work * deals in turn with the reliability, validity and efficiency of the method in evaluating the analgesic power of morphine and of some synthetic morphinomimetics : pethidine, l-methadone and levorphanol.

* The problem of the neuro-physiological mechanism of pain (121) and the manner in which narcotic analgesics act from the dual aspect of analgesia and addiction liability will be studied elsewhere.

Methods

We shall describe in turn the experimental conditions, apparatus, experimental procedure (125) and the methods of biometric analysis used.

I. Experimental Conditions

1. *Painful Stimulus*

Electrical stimulation was systematically used in our algesimetric study.

The most common types of current employed are faradic, sinusoidal and square-wave impulses and condenser discharges. Schroeder, 1907 (138); Heinroth, 1926 (61); Hesse & Reichelt, 1933 (66); Freund, 1936 (44); and Jores & Frees, 1937 (87) used faradic current. Goetzl et al., 1943 (50); Harris & Blockus, 1952 (57) and Fleisch & Dolivo, 1953 (41) preferred alternating current. Stimulation by condenser discharges was used by Koll & Reffert, 1938 (91) and by Kiesig-Orzechowski, 1941 (90). Finally, square-wave impulses were selected by Björn, 1946 (14), Soehring & Becher, 1949 (144), Stellmach et al., 1952 (149), Boréus & Sandberg, 1955 (17) and Yim et al., 1955 (178).

The parameters of the current (voltage duration and frequency of impulses), and the duration of stimulation and interval between stimulations vary with the type of current, the animal used, etc.

In most of the studies : Koll and Reffert, 1938 (91); Goetzl et al., 1943 (50); Stellmach, Schroeder & Soehring, 1952 (149); Fleisch & Dolivo, 1953 (41); Boréus & Sandberg, 1955 (17); Yim et al., 1955 (178), the voltage generally serves to define the threshold.

We used square-wave impulses with the following characteristics :

Frequency of impulses : 40 per second
Duration of impulse : 5 milliseconds
Voltage : variable
Length of stimulation : 0.5 sec.

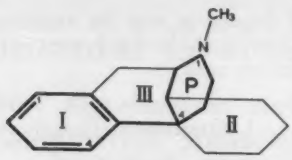
2. *Pain Receptors*

We selected the afferent dental nervous system as the pain receptor field. Stimulation can take place :

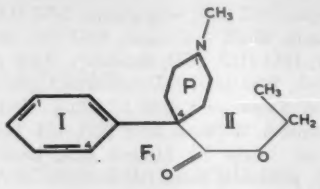
either, as indicated by Soehring (144), through the section of the dentine between the physical electrode and the tooth pulp, Steinhausen's " physiological electrode " (147);

or directly on the afferent nervous system in the dentine itself, the existence of such a system having been reported by Dependorf 1913 (31), Mummery 1924 (115), Münch 1927 (116) and Berkelbach 1934 (12). Recently Baud & Held, 1953 (8, 62), by an improved technique revealed the existence of sensitive fibres in the dentinal tubules in man. Baud, Radouco-Thomas & Nosal are now making a similar study on the guinea-pig's teeth. As to their nature, two types of sensitive and vegetative fibres have been described by Berkelbach 1934 (12), Brashear 1936 (19), Christensen 1940 (26) and Zerosi 1946 (180). Physiologically speaking, a sensation of pain is the afferent system's sole response to any kind of stimulus.

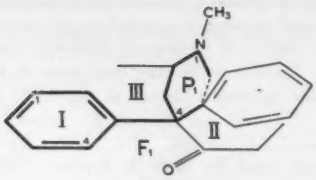
Two types of teeth have been used in electro-dental algesimetry—*Limited growth teeth*, such as the canine teeth of the cat (17) and dog. The stimulation of the dental receptors of dogs has been, and still is, one of the more available methods



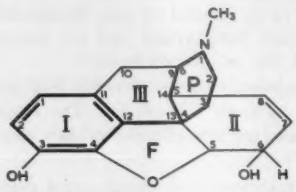
Methylmorphinan



Pethidine



Methadone



Morphine

FIGURE 1 — *Chemical structure of the synthetic analgesic narcotics studied*

for studying pain and analgesia. It is interesting to note that a similar technique has been used in man for some fifty years; Schroeder, 1907 (138), Heinroth, 1926 (61), Werz, 1932 (160), Hesse & Reichelt, 1933 (66) and Freund, 1936 (44), having used it for physiological and pharmacological research. Koll & Reffert first applied it in 1938 (91) to animal experiments to evaluate analgesic action, and it was subsequently used as such or with modifications by Kiesig-Orzechowski, 1941 (90), Goetzl et al., 1943 (50), Pfeiffer et al., 1948 (120), Soehring, 1949 (144), Fleisch & Dolivo, 1953, (41), Boréus & Sandberg, 1955 (17), etc.

Continuous growth teeth such as the upper incisors of rodents. This method, introduced by Gordonoff-Rückstuhl in 1939 (132) was used in the rabbit for pharmacological research by Wilhemi, 1949 (164), Domenjoz, 1952 (33), 1953 (34), Fleisch & Dolivo, 1953 (41), Frommel et al., 1953 (45), Sinniger, 1953 (142) and Yim et al., 1955 (178).

We have chosen as dental pain receptors the upper incisors of guinea-pigs. To our knowledge, there is no instance in medical literature of research on the dental organs of this species. The reasons for our choice are many. The guinea-pig gives to an electrical stimulus visible evidence of pain in the form of a clear-cut and constant biological response, similar to that of the dog. Furthermore, the response can be used for serial experiments, since the subject is a small laboratory animal, inexpensive and easy to handle.

We chose adult guinea-pigs of both sexes and average weight (500 g) living under standardized conditions (living space, food, temperature, etc.).

3. *Substances studied*

We studied the analgesic action of morphine and some synthetic morphinomimetics : pethidine, l-methadone and levorphanol (fig. 1). The doses are given in Table II (p. 29) and were administered hypodermically.

At the same time we studied the analgesic effect of some depressants of the central nervous system : Thiopental (pentothal) 10–30 mg/kg, Officinal ether, inhalated : 7 cc for 5 and 15 minutes in a bell-glass.

The doses given are expressed in milligrammes per kilo of animal weight (mg/kg). The effect of each dose was traced on 4 to 10 animals.

II. APPARATUS

The apparatus used for the algesimetric studies generally consists of a stimulating circuit and a few accessories.

1. *Stimulating Circuit*

This comprised a current generator and a system of electrodes.

(A) *The generator* may vary in type according to the form of current selected. Some authors (14, 41, 58) include a strong resistance in this circuit to counteract variations in the subject's resistance. For our experiments, we used a square-wave impulse generator—the Grass stimulator. An oscillograph was connected in parallel to check the stimuli.

(B) *The electrodes* constitute one of the most important elements in the apparatus, for on them depend the speed and accuracy of the method and the constancy of the results. Several types of electrode have been devised, varying in form, composition and above all, in mode of insertion.

In man, Schroeder, 1907 (138), Heinroth, 1926 (61), Hesse & Reichelt, 1933 (66), Freund, 1936 (44), Goetzl et al., 1943 (50), Björn, 1946 (14), Pfeiffer et al., 1948 (120) Sonnenschein and Ivy, 1949 (145), Harris & Blockus, 1952 (57) and Boréus et al., 1955 (17), used an active dental electrode and an indifferent electrode of the E.C.G. type.

For animal experiments two different types of electrode are used :

(a) *Fixed electrodes* for animals with limited-growth teeth. The prototype of this type of electrode, employed by various authors on dogs and cats, was evolved by Koll & Reffert in 1938 (91). They are of silver, inserted into a canine tooth, and connected by two insulated conductor wires the free ends of which are led through a facial fistula. They are connected in series to the stimulating circuit at the moment of stimulation. Soehring

et al., 1949 (144), Stellmach et al., 1952 (149) and recently Boréus et al., 1955 (17) made a number of improvements on this technique.

(b) *Removable electrodes*, used on animals with continuous growth teeth. For rabbits, the Fleisch & Dolivo's (41) type of electrode was generally adopted. This consists of an insulated clip supporting two pointed metal electrodes (0.8 mm in diameter) which fit into holes drilled in the incisors. On guinea-pigs we used a system of buccodental removable electrodes (figure 2).

The indifferent electrode. At the beginning of our experiments, a cutaneous indifferent electrode was fixed to one guinea-pig's fore-paws. After comparative study, however, we found it preferable to fix the indifferent electrode in the mouth. This ensured constant biophysical parameters and also prevented the animal from biting the research worker or dislodging the dental electrode with its tongue.

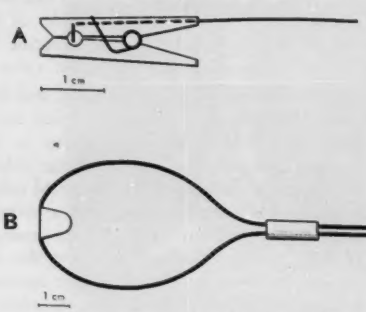


FIGURE 2. *Bucco-dental electrodes*

A. Active electrode (dental) B. Indifferent electrode (buccal)

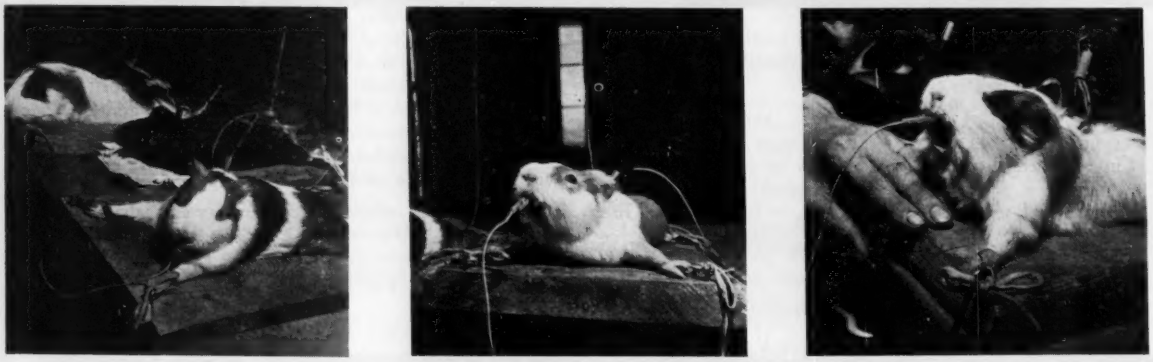


FIGURE 3. *Animal with the two electrodes inserted*

The bucal electrode consisted of a nickel-silver plate of about 1 sq cm soldered to a semi-rigid insulated wire which followed the line of the animal's jaw. This type of electrode has already been used by Radouco-Thomas et al. (121, 122) in a system of bucco-occipital electrodes for electrically induced epilepsy in animal experiments.

The active electrode. The active or monodental electrode is constituted by a 4/10 mm nickel-silver wire held by a microclip fixed perpendicularly on the upper incisors of the animal just before stimulation (figure 3).

The indifferent electrode remains in place throughout the experiment. The two electrodes are connected with the generator and the cathode-ray oscillograph. To permit of a serial experiment (nine animals) the wires to the electrode system are run from a bracket connected by a switch to the stimulating circuit.

Such electrodes are easy to devise and rapidly inserted (it takes about five minutes to insert the buccal electrode for a group of 6–9 animals) and remain very steady.

2. *Accessories*

These consist of a dental drill, boards for attaching the animals, etc. To perforate the teeth, we used an ordinary dentist's drill, but fitted with 0.5 mm drills. Larger drills (0.8 mm) of the type employed for drilling the teeth of rabbits were used at the beginning of our research, but these split the incisors, thus putting the animals out of action until the new teeth grew.

III. Experimental Procedure

The algesimetric experiments were in three phases :

1. *Preliminary Phase*

Our research showed that, to ensure a stable threshold voltage, it was preferable to drill the teeth the day before the experiment. The tooth selected must be sound, clean and long enough to bear a microclip (active electrode). The hole is drilled on the lateral upper side of one incisor about 0.5 mm below the gingival margin and to a depth of some 1/10 mm. The animal, unattached, is held still by the experimenter during the drill. Nine animals can be thus prepared in a quarter of an hour.

2. *Determination of the Pain-Response Threshold*

The animals are attached to boards, lying in " decubitus ventral ", and arranged in a row (6–9 animals) in a quiet room free from any sensitivo-sensorial stimuli. The insertion of the buccal electrodes is followed by some agitation. The guinea-pigs go through chewing motions and try to rid themselves of the electrode. After 10–15 minutes, however, they calm down and the threshold may be deter-

mined. The holes must be scrupulously cleaned before the pain measurements are made, as the tiniest particles of food affect the resistance of the tooth and cause an aspecific biological response or an abnormally high threshold value.

The threshold voltage is then determined by trial and error. All animals are first subjected to a voltage (3 volts) near to the average voltage ($\bar{x} = 3.16$). Then, according to their response they are subjected to rising or decreasing voltages until the pain-reaction test is obtained. The threshold voltage thus determined is checked twice by applying a series of three stimuli and repeating them after twenty minutes. The threshold grading in a series of animals saves the time that would otherwise be lost by observing the necessary interval of three minutes between each two stimulations. The determination is accurate to 0.1 volts.

3. *Evaluation of Analgesic Potency by the Increase of the Pain-response Threshold*

After grading, the animals receive the selected dose of the substance to be studied by whatever route has been chosen, and they are then subjected to increasing stimuli until the pain-reaction test is obtained.

The gradation of the stimuli for each animal is based on its control threshold voltage. The voltages selected were not in arithmetical progression (addition of a fixed number of volts or a percentage of the threshold voltage), but in geometrical progression : $\log V_{N+1} = \log V_N + k$. Each Excitation Unit (Ex. U.) represents an increase of 1/10 on the previous voltage:

Ex. U. = 0 : V_0 = Threshold voltage

Ex. U. = 1 : $V_1 = V_0 + V_0/10$

.

.

Ex. U. = N + 1 : $V_{N+1} = V_N + V_N/10$

The interval between Excitation Units 0,1,2, ... N is therefore smaller for low voltages than for high ones. This progression was chosen on the basis of Fechner's law of sensorial physiology : response = log (stimulus) + k. Whatever their control threshold voltage, the animals were sensitive to the interval chosen (1/10) and when stimulation was one Excitation Unit less than it should have been the result was always an incomplete response.

We therefore established a scale of supraliminal voltages for each animal derived from its threshold voltage by progression $V_{N+1} = V_N + V_N/10$, which the experimentor was bound to follow. This procedure adopted in conjunction with serial animal experiments with different threshold voltages, helps to counteract the subjectivity of the experimenter while he is working only on volts. We always observed an interval of three minutes between each stimulation and twenty minutes between each determination (three stimulations). If this time-table is not adhered to, too frequent stimulations cause an artificial rise in the threshold voltage.

The determinations were made in turn on two groups of four and five animals. This arrangement enabled measurements to be taken for each group in ten minutes.

Tests were made every thirty minutes or every hour according to the metabolism of the drugs, until the pain-reaction test was again forthcoming at the initial threshold voltage.

Each animal was used only once a week.

IV. Biometric Analysis of Results

The experiments with analgesics were mainly performed with three doses of three narcotic analgesics. To eliminate day-to-day variations (animals' diet, weather, ambient temperature), the nine animals in each pharmacological experiment were divided into groups of three, each of which received a different dose of the drugs. Each point on the graphs therefore represents the average of the results obtained under the same conditions (105, 106). The guinea-pigs, after administration of the drugs, were lined up at random to increase the objectivity of the experimenter in assessing the increase of the threshold (11, 159).

The analgesic effect was expressed in " Excitation Units " and not in volts, to eliminate the influence of the sensitivity of the normal animal to pain. Biometric analysis of the results was also made on the basis of these values. In the graphs reproduced in this document, we have added a scale showing the increase in the threshold value as a percentage of the initial threshold voltage, to permit of comparison with other data in medical literature.

The significant effect between doses was analysed by the " t " test (40, 104) :

$$t = \frac{\bar{x}_B - \bar{x}_A}{\sigma d \sqrt{\frac{1}{N_B} + \frac{1}{N_A}}}$$

Determination of the equiactive doses of the drug studied was based on the quantal response of the animal (39, 74). Analgesia was considered to have been induced in the animals when they showed no Pain-Reaction Test to a stimulus of 5 units (over 50% increase in the threshold).

Lethality was expressed in terms of the percentage kill in the groups of 5–10 animals receiving increasingly toxic doses of each product. The ED_{50} and LD_{50} doses were determined by the Miller-Tainter method. The margin of safety was fixed by the Index 50 and the therapeutical index (20, 107, 108).

Results

Our results are presented in two sections :

I. Study of the biological response to stimulation of the dental pain receptors in guinea-pigs;

II. Evaluation of the analgesic action.

I. Biological Reponse

1. Qualitative Study

Under the experimental conditions just described—i.e., taking as constant parameters a frequency of 40 impulses per sec., an impulse duration of 5 msec. and a stimulation duration of 0.5 sec. and subjecting the animal to increasing voltages—we obtained several types of response :

FIGURE 4

Reaction R_o (opening of the mouth)

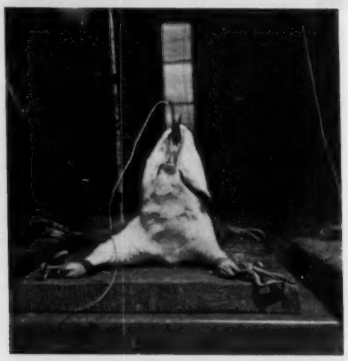


FIGURE 5

Pain Reaction Test (P.R.T. = R_1) front view (tied animal)

At a low voltage (less than 1 volt), the animal responded by a chewing movement or a slight tremor of the jaw, sensitive to the touch but difficult to see.

With stronger stimuli ($\bar{x} = 2.4$ volts $\pm$ 0.1), the animal responded by a slight, abrupt opening of the mouth (figure 4). This reaction, R_0, is constant, and its threshold regularly recorded.

Increases in voltage elicited an ever-wider opening of the mouth, subsequently accompanied by un-coordinated defensive movements on the part of the animal, as if it wished to rid itself of the tooth electrode.

At higher voltages ($\bar{x} = 3{,}16$ volts $\pm$ 0.06) a characteristic response (125) emerged which we have selected as the Pain-Reaction Test (R_1 = P.R.T). The main feature of this response was a rapid upward thrust of the head. Response is immediate, brisk and performed in a single smooth movement (figures 5 and 6).

In some animals, the angle of tilt was more than 90°, in others, less. With some the movement was accompanied by a contralateral or ipsilateral twist (fig. 7). These individual variations are extremely slight with tied animals, but quite definite with those in freedom. In conditions of freedom (fig 7*a*), changes in attitude were mainly as regards the intensity of the head movement. The upward thrust of the head is so violent that the animal loses balance and falls over backwards (figure 7*b*). We never observed any tendency to flight.

Although the response of the free animal is richer in physiological information, we performed all our experiments on tied animals. This position standardizes individual responses and makes it easier to differentiate subliminal responses (jerky upward thrust, in one or two jerks or undulating) from the Pain Reaction Test, and above all makes it possible to perform experiments in series.

At even higher voltages, the animal's response was of the same type, but more violent and often accompanied by a cry.

At very high voltages (about 10 volts), the upward thrust of the head disappeared completely and was replaced by a " horrified " recoil of the head, accompanied by a cry (R_2). Mydriasis, a certain bristling of the hair and trembling of the

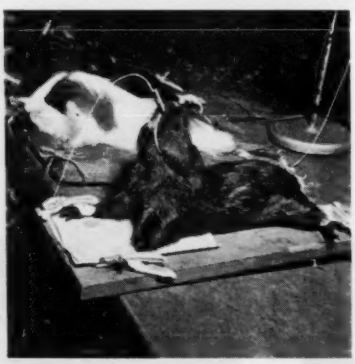


FIGURE 6

Pain Reaction Test, profile (tied animal)

FIGURE 7

Pain Reaction Test, contralateral (tied animal)

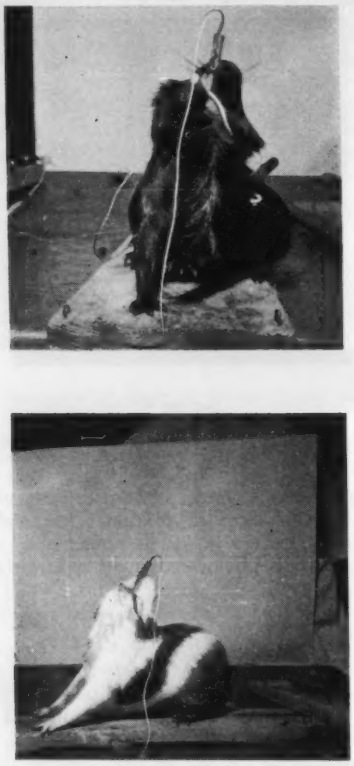


FIGURE 7*a*

Pain Reaction Test, free animal, front view and profile

hind-quarters were also noted. This stimulus was followed by a few groans and a state of excitation. In free animals, the same increase in voltage elicits first an agitated movement of recoil and then flight.

Such were the various aspects of the somatic responses obtained from guinea-pigs by electro-dental stimulation.

The extent to which they can be regarded as an objectivation of the sensation of pain in guinea-pigs following dental stimulation will be considered in the discussion chapter.

Of the various responses, we chose the single upward thrust of the head as the Pain Reaction Test (PR.T) owing to its clear-cut nature, regularity and reproducibility. This choice, as we shall see later, was also justified by a number of neuro-physiological and pharmacological criteria.

Before passing to the quantitative study of the P.R.T., it may be advisable to give some information on the much-debated question of *training* the animals.

No training period was required to obtain the P.R.T. with guinea-pigs. There were no variations between the first and later determinations. At the threshold voltage, the animals showed the same type of P.R.T. in the various series of experiments.

With animals already subjected to the test, the threshold voltage appeared, however, to be easier to determine than with new animals, the upward thrust of the head attaining more swiftly a stable threshold with a characteristically clean movement.

The only factor requiring training is the experimenter, who has to become used to recognizing the P.R.T. and distinguishing it from subliminal responses (jerky or undulating movement) and supraliminary responses (more violent movements).

It should, however, be pointed out that during the various sessions to determine the control values prior to each pharmacological experiment, we sometimes obtained for one or other of the animals a regular non-typical response or a response of type R_2, whatever the voltage used. These animals were excluded from the current experiment, but gave a typical P.R.T. again on following days or after a new drilling. Such unexpected and passing deficiency appears to have been due to some anomaly in the conditions of the experiment.

Subsequently we avoided this drawback by drilling holes the day before in two incisors. Then, if the stimulation of one tooth brought a non-typical response or one of type

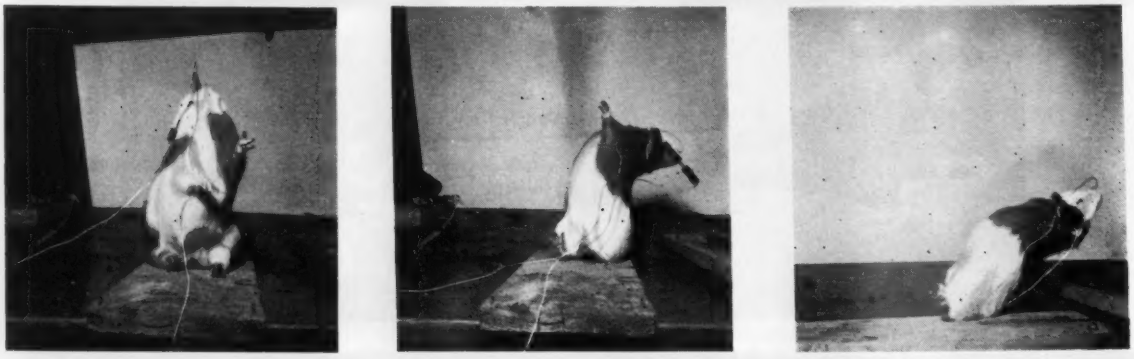


FIGURE 7*b*

Pain Reaction Test, free animal. The intensity of the reaction is such that the animal loses balance

R_2, it was enough to stimulate the other incisor in order to obtain a typical P.R.T. The animal could then be used for the experiment.

2. Quantitative Study

Under this heading, we analysed the P.R.T. and, occasionally, responses R_0 and R_2. The lowest voltage eliciting a characteristic P.R.T. was taken as the threshold voltage or threshold for reaction.

We will now present the variations in threshold voltages in their relation first to the animal and then to the parameters of the stimulus.

A. *Threshold Voltage and Biological Factors*

(a) *Inter-animal Variations*

The study of inter-animal variations mainly concerns the Pain Reaction Test. A series of supplementary measurements were made of the R_0 response. Figure 8 gives a block diagram of the P.R.T. threshold voltages for 221 determinations and shows that guinea-pigs have an average threshold voltage of 3.16 volts $\pm$ 0.06.

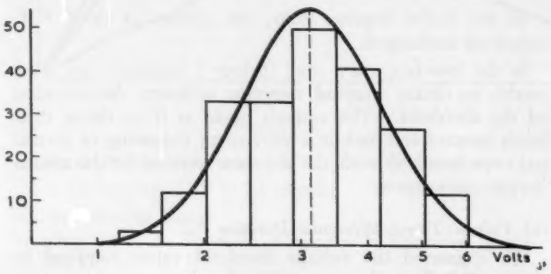


FIGURE 8

Block diagram of the threshold voltage of the Pain Reaction Test
x axis : voltage (logarithmic scale)
y axis : number of animals

In 167 cases we also registered the voltages for R_0 and R_1 during the same experiment. These are shown in the double block diagram in Figure 9. It will be seen that the average voltage for R_1 shows an increase of 50% over that for R_0.

$$x_{R_0} = 2,4 \pm 0,1 \qquad \bar{x}_{R_1} = 3,7 \pm 0,1$$

This ratio, regularly encountered when establishing the individual control values, was confirmed by statistical study,

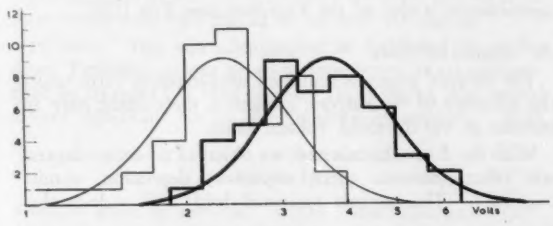


FIGURE 9

Block diagram of the threshold voltages of R_0 *and* R_1 (P.R.T.)
x axis : voltage (logarithmic scale)
y axis : number of animals

the correlation coefficient " r " having the highly significant value of 0.879.

(b) *Intra-animal Variations*

We studied the variations of the threshold voltage in the same animal on the same day with one drill-hole and with several drill-holes.

On any single day, the response threshold of the animals proved remarkably constant.

During the pharmacological experiments, the control animals were subjected for the same time (3–7 hours) to stimulation at the same rate as the drugged animals. Their thresholds proved constant (figure 20), a subliminal and a superliminal stimulus ($\pm$ 1 stimulation unit) eliciting respectively a subliminal and a superliminal response.

We also obtained a consistent return to the initial threshold voltage with drugged animals. This stability of the threshold of response in the course of a single experiment is the basis for the pharmacological study of analgesic action. As pointed out in the section on method, the increases in the threshold value recorded in all the pharmacological experiments were always related to the individual premedication threshold voltages.

Thus each point represents not the average increase in the voltage, but the average of the elevations of the individual threshold voltages. This explains why, despite inter-animal variations in the threshold voltage, all the curves (figures 13, 14, 15 and 20) have a common point of departure, V_0, expressing all the initial threshold voltages.

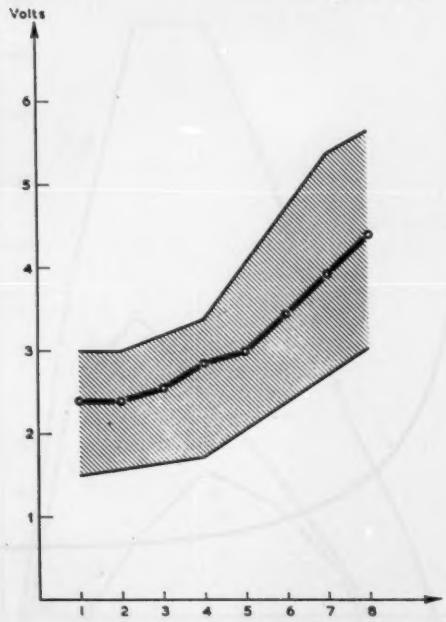


FIGURE 10

Variations of the threshold voltage of the Pain Reaction Test during one week for one drilling (4 animals)
x axis : time in days
y axis : voltage

As a matter of interest we also recorded *variations in the threshold voltage during the days following the drilling.* For the first two or three days it remained constant. After this, however, there was a steady rise in the threshold voltage, inversely proportional to the downward movement of the hole (figure 10). Any given hole could be used only during the 8 to 10 days after drilling, owing to its gradually moving farther away from the tooth pulp and to the increasing difficulty of fitting the micro-clip.

The variations in the threshold voltage in the same animal from one drill to another are quite unrelated.

For example, the following variations were registered on one animal with 8 perforations : 2.5, 2.7, 3.4, 3.6, 2.9, 2.5, 1.4, 2.4 volts. This variability of threshold voltages in the same animal brings out the fact that the threshold voltage value depends more on the electrode-receptor complex than on the threshold of the animals' sensitivity to pain.

B. *Threshold Voltage and Biophysical Factors* (parameters of the current)

We studied the variations in the threshold voltage of the P.R.T. in relation to the frequency and duration of impulses.

(a) *Threshold voltage/impulse Frequency Ratio*

With a constant impulse duration of 5 m.sec., we subjected five animals to frequencies varying from 7.5 to 100 imp/sec. The threshold voltages required to elicit the P.R.T. under such conditions are shown in figure 11.

It will be seen that the voltage/frequency ratio values can be plotted as a hyperbola

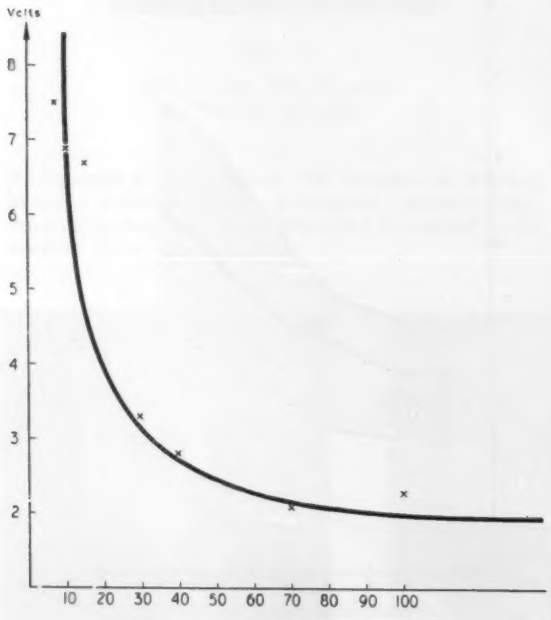


FIGURE 11

Threshold voltage/Impulse frequency ratio

x axis : impulse frequency

y axis : voltage

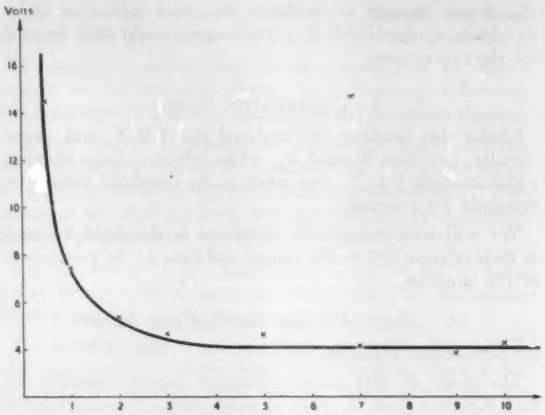


FIGURE 12. *Threshold voltage/Impulse duration ratio*

x axis : impulse duration (msec.)

y axis : voltage

In the 9–100 imp/sec. zone, the quality of the P.R.T. remained unchanged.

In the low-frequency zone (below 9 imp/sec.), we were unable to obtain a typical response, whatever the elevation of the threshold. The animals made as if to thrust their heads upward and back in a movement consisting of several jerks synchronized with the impulses received by the animal during stimulation.

(b) *Voltage Threshold/Impulse Duration Ratio*

We registered the voltage threshold values required to elicit a P.R.T. with various impulse durations at constant frequency (40 imp/sec). A typical response was obtained only for durations from 0.2 to 7 m.sec. The points calculated for five animals can be plotted as a hyperbola (figure 12).

II. Evaluation of Analgesic Action

The results obtained for morphine and three synthetic narcotic analgesics, pethidine, *l*-methadone and levorphanol, are given below. For purposes of comparison, a few results are appended for two central nervous system depressants, ether and thiopental.

1. *Narcotic Analgesics*

We will consider the effect of the narcotic analgesics first on the form (qualitative study) and then on the threshold (quantitative study) of the Pain-Reaction Test (125).

A. *Qualitative Study*

The animals' response remained unchanged in form under the influence of the narcotic analgesics, there being only an increase in the threshold voltage value.

With the doses administered, we observed no excito-depressive effects (narcotic effect, respiratory depression, vomiting, etc.). The animals remained brisk and wide-awake.

B. *Quantitative Study*

We first studied, substance by substance, the intensity and duration of the analgesic action with increasing doses (dose/effect/time ratio).

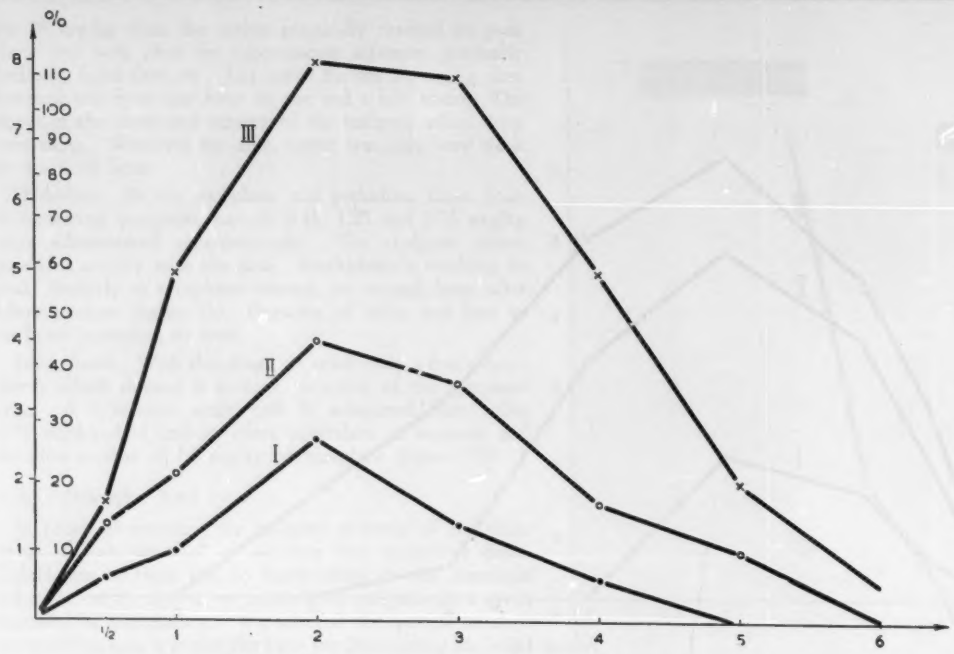


FIGURE 13

Analgesic action of morphine. Curve : dose/effect/time ratio :
I = 0.8 mgr/kg II = 2.5 mgr/kg III = 7.5 mgr/kg
x axis : time in hours
y axis : rise in the threshold of response in Excitation Units (Ex.U.) and in %

We then made a comparative study in connexion with the dose/effect ratio (percentage of animals showing analgesia or percentage killed) of the analgesic effect and the margin of safety of the drugs.

(a) *Dose/effect/time Ratio*

Morphine. Three doses, in a ratio of 1/3 (0.83 mg/kg, 2.50 mg/kg and 7.50 mg/kg), were administered to the guinea-pigs by subcutaneous injection. A significant effect was already observed with the 0.83 mg/kg dose. Whatever the dose, the analgesic action developed gradually, reaching its peak towards the second hour after administration (figure 13).

The duration of effect increased with the dose, varying from three hours with 0.83 mg/kg to six with 7.5 mg/kg.

Pethidine. This was administered in 3, 10 and 20 mg/kg doses. Pethidine, as can be seen from figure 14, is characterised by its rapid but brief action. The 3 mg/kg dose caused a very significant rise in the threshold of response. With

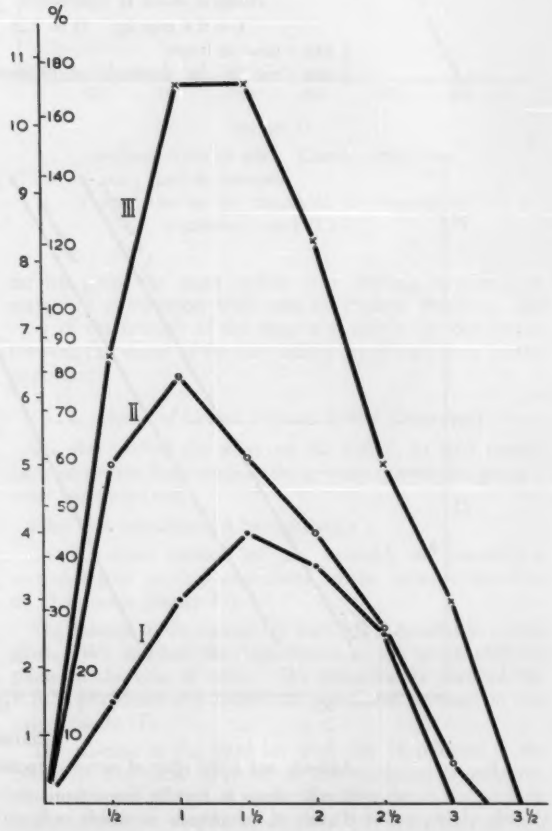


FIGURE 14

Analgesic action of pethidine. Curve : dose/effect/time ratio :
I = 3 mgr/kg II = 10 mgr/kg III = 20 mgr/kg
x axis : time in hours
y axis : rise in the threshold of response in Excitation Units (Ex.U.) and in %

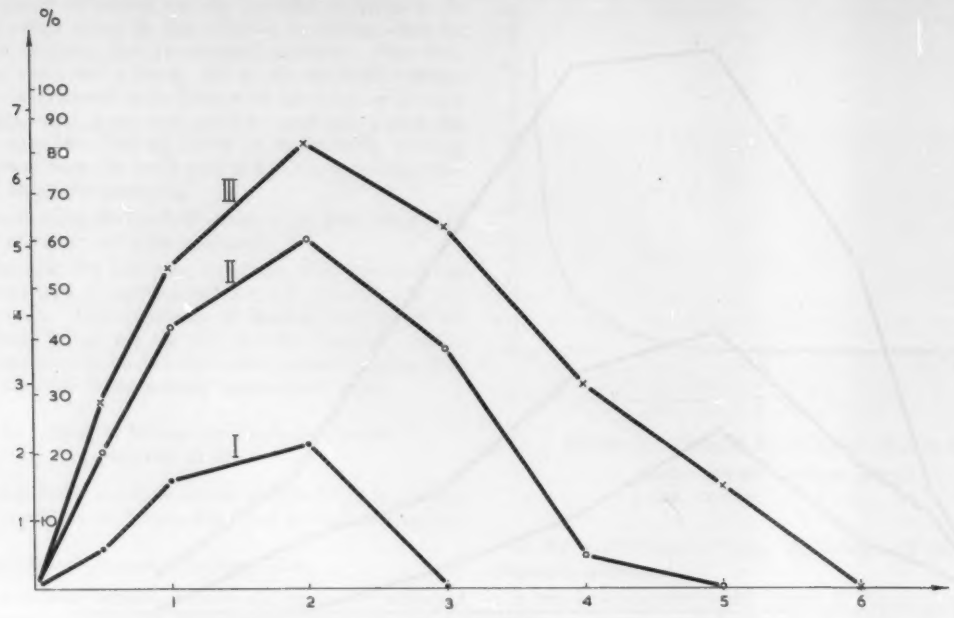


FIGURE 15

Analgesic action of l-*methadone*. Curve : dose/effect/time ratio :
I = 0.4 mgr/kg II = 1.25 mgr/kg III = 3.75 mgr/kg
x axis : time in hours
y axis : rise in the threshold of response in Excitation Units (Ex.U.) and in %

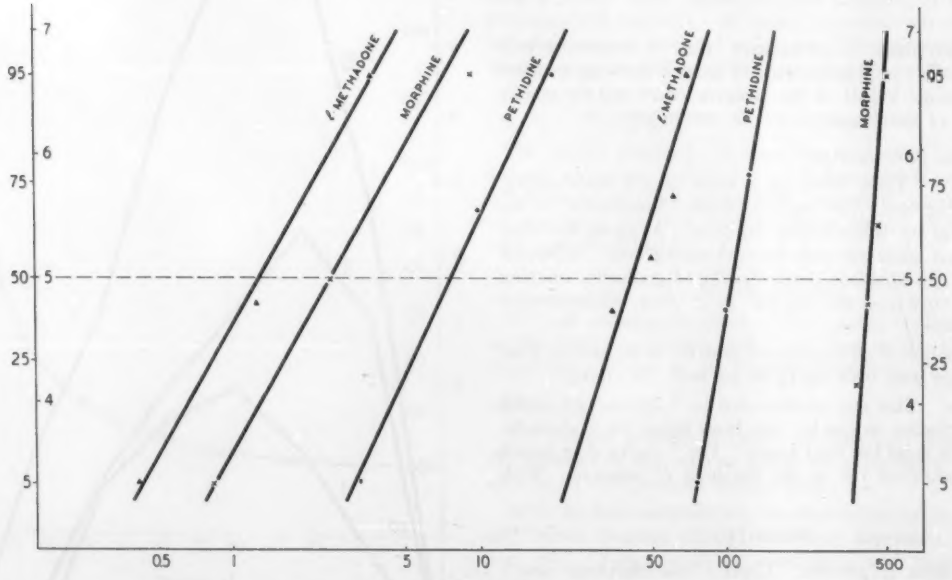


FIGURE 16

Analgesic and lethal effect of morphine, pethidine and l-*methadone*. Curve : dose/effect
x axis : doses in mgr/kg (logarithmic scale)
y axis : % of animals in which analgesia was induced; or dead animals (probits)

the 10 mg/kg dose, the action practically reached its peak about one hour after the subcutaneous injection, gradually declining from then on. The curve for the 20 mg/kg dose flattened out from one hour to one and a half hours. The slopes of the onset and waning of the analgesic effect were very steep. Whatever the dose, action was only very weak in the third hour.

Methadone. As for morphine and pethidine, three doses in increasing quantities, namely 0.41, 1.25 and 3.75 mg/kg were administered subcutaneously. The analgesic action increased steadily with the dose. Methadone is reaching its peak similarly to morphine towards the second hour after administration (figure 15). Duration of effect was four to six hours according to dose.

Levorphanol. With this drug, we made only a few experiments which showed it to have an action of the morphine type. A systematic study will be submitted later. The 3.75 mg/kg dose had an effect equivalent in intensity and duration to that of 7.5 mg/kg of morphine (figure 20).

(b). *Dose/effect Ratio*

In order to compare the analgesic potency of the three drugs studied, we tried to ascertain their equiactive doses. Calculation of these can be based either on the threshold value, or on the absence or presence of analgesia for a given intensity of stimulation. We selected the quantal method for preference, as it is also the basis for determining the lethal dose and thus permits of subsequent comparison of zones of activity and lethality.

Lots of 5–10 animals, each given progressively larger doses of the analgesics under study, were subjected during the hour of peak effect of each product to a standard stimulus representing five stimulation units (an increase of 50 per cent over the control threshold voltage). We noted the percentage of animals in each lot in which analgesia was induced—i.e., those not giving the Pain Reaction Test.

The straight-line curves delimiting the zone of activity of each product, are shown in figure 16.

The doses inducing analgesia in 50 per cent of the animals (analgesic dose 50 = ED_{50}), calculated by the Miller-Tainter method (113) are given in table I.

We also determined the toxicity of the same products in guinea-pigs. Once the $analgesic_{50}$ and $lethal_{50}$ doses are known, one can work out the corresponding $index_{50}$. These indices (table I) illustrate the very broad margin of safety in the guinea-pig, between the analgesic and the lethal effect of the narcotic analgesics. The analgesic action does not interfere with the toxic side-effects either. This question will be dealt with again during the discussion.

TABLE I

Doses		*Morphine*	*Pethidine*	*Methadone*
Dose $_{50}$ in mg/kg	ED_{50}	2.5	7.7	1.3
	LD_{50}	379	106	41
Index $_{50}$		151	13.7	31
Therapeutical index		44	5	5.7

The straight-line curves of effect and toxicity not being parallel, the therapeutical index is a more accurate means of estimating the margin of safety (127). We have none the less given the more widely used $Index_{50}$ in table I to permit of comparison with data in medical literature. In view of the breadth of the margin of safety, the differences between the values of the two indices are of only very minor importance.

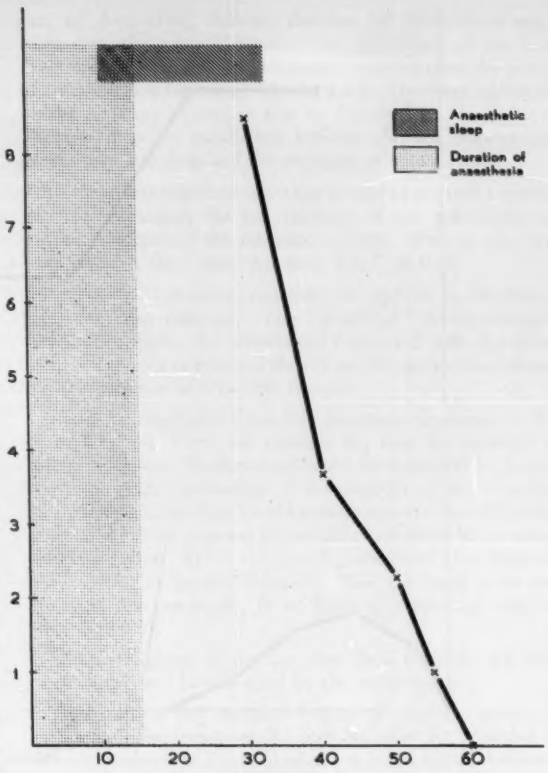


FIGURE 17

Analgesic action of ether. Curve : effect/time
x axis : time in minutes
y axis : rise in the threshold of response in excitation units (Ex.U.)

2. *Action of Central Nervous System Depressants*

We also studied the effect on the P.R.T. of two central nervous system depressants in the general anaesthetics group : ether and thiopental.

Ether was administered by inhalation :

On an initial control lot (six animals), we recorded a second-degree general anaesthesia of an average duration of 35 minutes (figure 17).

The animals in the second lot were left 5 minutes in a bell-glass. We stopped the experiment in the pre-anaesthesia phase in the case of ether. We immediately checked the P.R.T. threshold and found no significant increase in the value (figure 17).

The animals in the third lot were left 15 minutes in the bell-glass and then subjected to electro-dental stimulation. The results are plotted in figure 17. This shows that as long as the anaesthetic sleep lasts, the P.R.T. is completely absent;

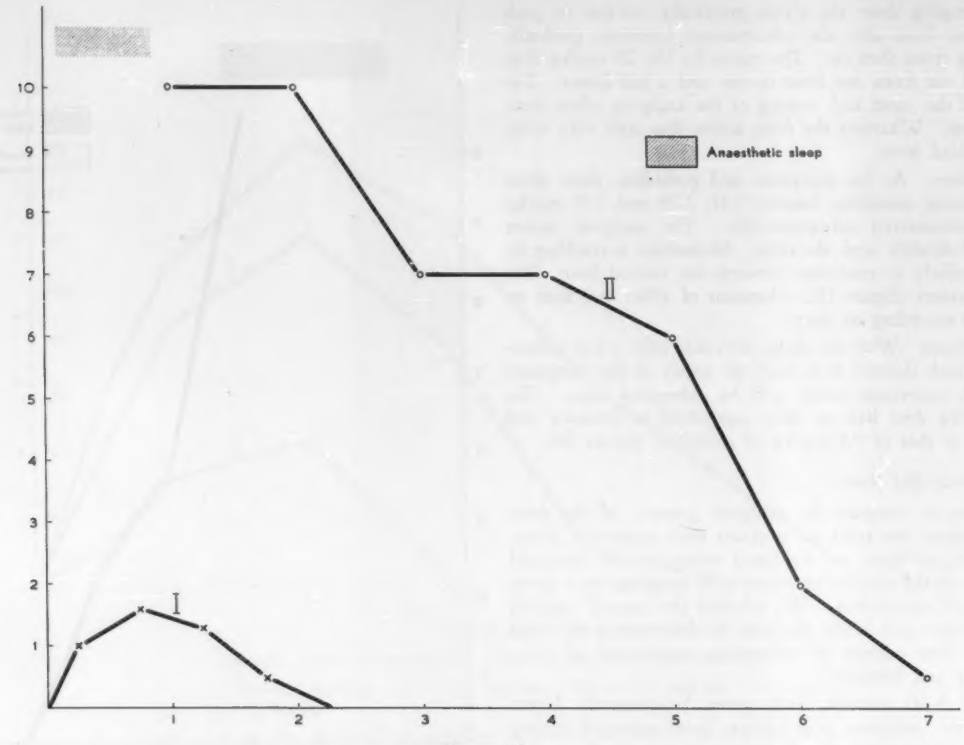


FIGURE 18

Analgesic action of thiopental. Curve : dose/effect/time ratio : I = 10 mgr II = 25 mgr
x axis : time in hours
y axis : rise in the threshold of response in excitation units (EX.U.)

even very high voltages (about 20 volts) provoke only a R_0 type reaction.

On waking, the animals at first give a P.R.T. only at very high voltages (120%). The response returns to the initial threshold after about 50 minutes.

Parallel with the elevation of the threshold, there is a change in the quality of the P.R.T. It loses its vigour, becoming feeble and sluggish.

In the six animals studied, administration of *thiopental* in doses of 10 mg/kg induced sedation accompanied by a slight increase in the threshold value (about 15%), for an average duration of 60 minutes. The response, as in the case of ether, was feeble and sluggish. Return to normal was gradual and took a few hours.

With a hypnotic dose, 25 mg/kg (on four animals) it was, as in the case of ether, impossible to obtain a P.R.T. a long as the anaesthetic sleep lasts. The initial threshold value was returned to after about 7 hours (figure 18).

Discussion

For an algesimetric method to be used in experimental medicine and pharmacology, it must meet a series of basic requirements with respect to reliability validity and efficiency.

I. Reliability of the Method

For the method to be reliable :

(1) It must be certain to elicit in the animal a sensation of pain which can be identified by physiological reactions (somato-vegetativo-humoral).

(2) The responses must be easily and clearly definable, qualitatively speaking, and capable of quantitative evaluation.

(3) It must be able to demonstrate the analgesic action of substances of established clinical efficacy.

We will first discuss the physiological and then the pharmacological data which ensure the reliability of the method.

1. *Physiological Data*

There are as yet no methods of neuro-physiological investigation of sufficient accuracy for the direct evaluation of pain phenomena. As Jacob, 1954 (82), points out, the most objective biological response would be the electrical disturbances recorded at the centres and along the path of the pain. Little work has been done in this field : Nims et al. 1941 (118), Andrews 1942 (3), 1943 (5), Cahen & Wikler, 1944 (23), Leimdorfer 1948 (97), Toman & Davis, 1949 (157), Wikler 1950 (162), Scherrer et al., 1954 (134), 1955 (135), and Monnier & Gangloff, 1955 (49), 1956 (114b). Such research is just beginning.

For this reason, algesimetric studies have still to be made through the medium of indirect biological responses. In man, information on the spontaneous (pathological) or induced (experimental) sensation of pain is obtained from the patient's own description.

In animal experiments, a series of somatic reactions of the quantal type may be said to provide evidence of the presence and intensity of the sensation of pain. In the guinea-pig, we induced by progressive electric stimulation of the dental receptors a series of reactions (R_0, R_1, R_2) described in the chapter on results.

A. *Are the Reactions Evidence of Pain?*

The fundamental problem which arose before the analgesics could be studied was whether the reactions were really the visible expression of a pain phenomenon and, if so, whether the vigour of the pain reactions increased regularly in proportion to the intensity of the stimulus.

Response R_0.—Response R_0, opening of the mouth (figure 4), is characterized by its uniformity and monotony. Is it a response to a minimum sensation of pain or a simple reflex without pain?

Pfeiffer et al., 1948 (120) report that it is possible to induce in man by dental stimulation a faint pain with a very low threshold (T_1) and a severe pain with a higher threshold (T_2). According to Harris and Blockus, 1952 (57), the first sensation is not painful, and it is only when the stimulus becomes more intense that a clear sensation of pain emerges. Koll et al., 1938 (91), record the emergence of uniform reactions at low voltages : muscular jerks in the floor of the mouth, licking movement of the lip on the side of the stimulated tooth, but do not regard these as pain-responses. Soehring and Becher, 1949 (144), describe a response, syndrome Z, in dogs, similar to R_0 in the guinea-pig, which is constant and monotonous in form and development. They regard it as a mere reflex representing the earlier stage in the motor effect provoked by the dental stimulus.

Response R_1 *(P.R.T.).*—The qualitative study shed some light on the concrete evidence of pain furnished by the testing reaction. Although this reaction takes the form of a primary movement—a clear-cut and easily identifiable single upward thrust of the head as far as it will go (figures 5 and 6)—each animal shows individual secondary variations. Moreover, there is often a cry at the threshold voltage or, more often, at superliminal voltages. The individual characteristics, more marked when the animal is free, are probably, as reported by Koll & Reffert in the case of dogs, the sign of a pain-response and not of a simple reflex without pain.

The quantitative study, based on variations in the frequency and duration of the impulses of the stimulus, show, as Soehring observed in the case of dogs (144), that the test response of the guinea-pig may be regarded as a pain reaction. The voltage/frequency ratio can be graphically represented as a hyperbola (figure 11), which should be an indication of a summation phenomenon. This phenomenon, studied by Magun, 1948 (110), is said to be characteristic of the pain sensation in man. Scherrer et al., 1954 (134), 1956 (136), have recently made an important contribution to study of the pain sensation phenomenon.

Similarly, the voltage duration of stimulus ratio in the guinea-pig can be expressed as a hyperbola (fig. 12), thereby indicating that the threshold of the test reaction, as in the case of dogs (144), follows the law of Weiss-Hoorweg. Moreover, the relation between the emergence of the test reaction and the quantity of electricity required takes the form of a straight-line function. Similar results have been obtained on the sensation of pain in man as described by the patient. There is therefore parallelism between the test response in guinea-pigs and dogs and the sensation of pain in man.

All these data together may thus be said to provide a sound basis for regarding the test reaction of the guinea-pig as somatic evidence of the sensation of pain. That is why we have called it the " Pain Reaction Test " (P.R.T).

Response R_2.—A pain syndrome also appears to be clearly present in this response. The " horrified " recoil, struggle and sudden flight, the phenomena connected with the sympathetic nervous system and the cry are all symptoms indicating the presence of a painful stimulus.

It is to be concluded from this discussion that response R_1 (Pain Reaction Test) and response R_2 may be regarded as reactions to pain. Furthermore these reactions seem to present responses graded according to the intensity of the stimulus. The Pain Reaction Test would correspond to a bearable pain, whereas the flight response R_2 would be evidence of excessive, paroxystic pain. As for response R_0, we can neither disprove nor confirm its painful character. We can only point out the close relation R_0/R_1 ($r = 0.86$) in a constant ratio of 50% (figure 9).

Additional proof of the fact that these reactions are alike in character will be provided by the analgesia tests.

Thus, within the complex biological response system of the guinea-pig, reactions R_1 and R_2 may be regarded as somatic evidence of a painful sensation, a " sichtbare Ausdruck empfundenen Schmerzes " (144). For analgesimetric studies we have kept the Pain Reaction Test (P.R.T. = R_1), which lends itself to both qualitative and quantitative study.

Along what paths does the Pain Reaction Test travel and what is their relation to the pain centres?

According to Soehring & Becher (144), Pain Reaction Test and the syndrome R in dogs are defensive movements controlled by the diencephalon and the brain stem. Great similarity has been found by these authors between the test response in dogs and the affective defence reaction (" Affektive Abwehr-reaction ") of Hess & Magnus, 1943 (64, 65) elicited from cats by excitation of the diencephalon. The experiments of Amsler, 1921 (2) and Keller, 1932 (89) on thalamic animals seem to support the same view. One objection may be raised—namely, that in the tooth stimulation of cats Boréus could obtain no reaction similar to the complex defensive movement. Be that as it may, the correlation reported by Soehring is really convincing and offers a very interesting working hypothesis for new research.

On the basis of these data and the results set out under the heading of biological response, the P.R.T. of the guinea-pig may be regarded as a long supraspinal circuit. Our results with analgesia and the work of Winder et al., 1946 (165) and 1947 (166), Houde & Wikler, 1951 (71), Irwin & Houde, 1951 (75), Winter & Flataker, 1953 (169*a*) and Jacob, 1954 (82) on defensive reactions to nociceptive thermal stimuli would confirm this view.

Research is in progress on these circuits and their exact relation to the pain centres. The results will be published later (128).

B. *Qualitative and Quantitative Stability of the P.R.T.*

As already pointed out, for an algesimetric method to be reliable the pain-reaction selected as the test response must be capable of qualitative definition and of measurement.

The qualitative study of the P.R.T. of the guinea-pig—single upward thrust of the head as far as it will go accompanied by some minor individual vatriations—shows that response to be clear, easily identifiable, steady and reproducible in that form.

It may be of value to compare the biological response obtained by stimulating the tooth pain-receptors of other animals. We have found a biological response similar to the Pain Reaction Test of the guinea-pig only in work on dogs. This reaction was considered as characteristic of a highly organized animal (91). Its presence in the guinea-pig seems to suggest that it is not confined to any animal species and is quite likely to appear in other laboratory animals (cats or rabbits) under suitable experimental conditions.

Even in dogs it does not always appear. Koll et al. (91, 92, 93) found it with its qualitative and quantitative characteristics in all the animals studied. Soehring and al. (144, 148, 149) did not encounter it regularly. It may be replaced by other types of response (Z syndrome and R syndrome). Wirth (170) working on dogs, managed to single out four, which gave constant threshold values for a year. One dog developed a conditioned reflex, giving the test response merely on hearing sound made by the apparatus. Boréus and Sandberg (17) succeeded in eliciting the Koll reaction consistently. But they had to select the animals somewhat, as with some dogs the reaction was difficult to recognize, while with others there were day-to-day variations of more than 15 volts in the threshold value.

The responses of the R_0 and R_2 type only, which we sometimes encountered (c.f. chapter on method) in one or more of the animals tested and which gave way after a new drilling to a typical P.R.T. might perhaps provide a partial explanation for the exceptions found among dogs. Through the use of removable electrodes with guinea-pigs it was possible to show that the absence of the test response was purely apparent, but due to the experimental conditions.

There is another problem which can be raised. In guinea-pigs the Pain Reaction Test was from the beginning constant in appearance and could be reproduced without any training. Koll and Reffert (91) report that, in dogs, the " Reaction " appeared from the beginning with its qualitative and quantitative characteristics. Soehring and Becher (144), on the other hand, report that the characteristic biological response developed only in the course of the experiments. Similarly, Boréus and Sandberg (17) claim that 4–5 weeks' training is necessary to obtain a steady response.

The biological response in rabbits after dental stimulation is characterized by a chewing movement (41, 132). Being easy to reproduce and constant in form, it tends sometimes to become a conditioned reflex. As for the somatic reactions of the cat, qualitative defects make them of little use in algesimetry (17).

From the quantitative standpoint, the reliability of the tooth-pulp algesimetric method with guinea-pigs is ensured by the stability of the potential stimulus and, above all, of the actual stimulus.

Use of electrical stimulation and the apparatus described permits of accurate measurement and reproduction of the parameters of the potential stimulus. Although with guinea-pigs continuous growth type teeth involved the use of removable physical electrodes (figure 2), we were able to obtain a physiological electrode (stimulus-receptor complex) of great stability. This stability of the actual stimulus is reflected in the remarkable steadiness of the Pain Response Threshold (threshold voltage) in any particular experiment (figure 20).

The progressive increase in the threshold value in the days following the hole drilling indicate that the stimulus-receptor contact is becoming steadily weaker. Whatever its level, however, the threshold voltage always stays constant during the day of the experiment. Physiological researches and pharmacological experiments with substances of rapid metabolism (thiopental) could thus be made on the same animal for a whole week after drilling the hole.

The slight dispersion of individual response thresholds and their distribution along a Gauss curve (figure 8) may be taken as an indication of a fairly uniform physiological electrode and of slight variability in the thresholds of sensitivity to pain (receptor-pain centres complex) in the various animals. For 221 gradings, we have had an average threshold voltage of 3.16 ± 0.06 volts. The slightness of the dispersion facilitates grading.

Koll & Reffert (91), with fixed electrodes, obtained a constant threshold voltage throughout several months of testing. With certain dogs, the value was absolutely steady, but with others some variations in threshold were recorded. They are of the same order of magnitude (32–40 volts) as the inter-individual variations recorded with four dogs. Boréus and Sandberg (17) recorded day-to-day variations of more than 15 volts in the threshold value with certain dogs. With rabbits and removable electrodes, Fleish & Dolivo obtained an average threshold voltage of 18 volts with a standard error of 7.5 volts, in the course of 1,600 experiments.

Inter-animal variations, in any case, are of only relative importance. Even experimenters working with fixed electrodes in practice carry out grading before each pharmacological experiment. It therefore follows that the reliability of algesimetric results is ensured by extremely accurate control determination coupled with strict constancy of the threshold voltage during the experiment.

We have described the physiological condition ensuring the reliability of the algesimetric method using the tooth pulp of guinea-pigs—namely, the presence of a qualitatively and quantitatively identifiable response as evidence of pain. Let us now consider the degree of reliability of the method from the pharmacological standpoint in the light of the results obtained with narcotic analgesics.*

2. *Pharmacological Data*

A. *Reliability of the Method*

Since one of the main objects of any algesimetric method is to evaluate the analgesic action of new products, it is essential first to check its reliability by studying drugs whose analgesic action has been confirmed by clinical experience. We will therefore consider in turn the results relating to the analgesic action of morphine, pethidine, methadone and levorphanol (figures 13, 14, 15 and 20).

* The effect of antipyretic analgesics has been studied at greater length elsewhere (125). Let us merely mention that the action of 100 mg/kg of Aminopyrine and of 100 mg/kg of Butazolidine is similar in intensity to that of 2.5 mg/kg of morphine but is of longer duration (about 5 hours).

TABLE II

Drug ⟶			Morphine			Pethidine			l-Methadone		
Dose in mg/kg ⟶			0.8	2.5	7.5	3.3	10	20	0.4	1.25	3.75
Elevation of threshold		$\bar{x}$	2	5.26	7	3	6.3	10.6	2.6	4	8
	Excitation Units	St. E of $\bar{x}$	± 0.94	± 0.8	± 0.8	0	± 0.5	± 0.7	± 0.5	± 0.6	± 1.1
	% of threshold voltage	$\bar{x}$	22.3	65	87	33	83	178	29	48	119

The rise in the threshold of the Pain Reaction Test brought about by increasing the dose of the drugs provides a means of checking the sensitivity of the method and its selectivity in two main respects, intensity and duration.

The small doses required to obtain a minimum of analgesia bring out the *sensitivity* of the method. For the three drugs studied, there was a significant rise in the pain threshold with such tiny doses as 0.4 mg/kg for methadone and 0.8 mg/kg for morphine. With 3 mg/kg of pethidine a highly significant effect was obtained.

B. *Selectivity of the Method*

To study the *selectivity* of the method, we have grouped in table II below values showing the intensity of the analgesic effect obtained by increasing doses of the three drugs at peak effect period—i.e., the second hour in the case of morphine and *l*-methadone, and the first hour in the case of pethidine.

The intensity of the analgesic effect is expressed in terms of Excitation Units ($\bar{x}$ and σ) and of the percentage rise in the threshold valul (x).

It will be seen that the elevation of the threshold is progressive and proportioned to the dose.

Figure 19, giving the above data in graphic form, shows a linear relation between the increase in dose and the analgesic effect, expressed in excitation units (log. of the stimulus). The progression did not reach a ceiling in the zone studied.

Application of the " t " test enabled us to check whether the ratios of 1 : 3 and 1 : 2 chosen for the increase in dose were sufficient to produce significant elevations of the threshold value (analgesic action). As table III shows, significant figures are obtained for methadone and highly significant ones for pethidine. In the case of morphine, it seems that it would have been preferable to have had a broader spread of doses. However, the difference between the doses is significant if comparison is made not at the point of peak effect, but over the whole of each curve (effect/time ratio).

The determination of the equiactive doses (figure 16) based on the percentage of animals showing analgesia (i.e., showing over 50 per cent increase in the threshold value) enabled us to evaluate the relative potency of the drugs studied (figure 16). If morphine is taken as the reference drug (ED_{50} = 2.5 mg = 1), the relative potency of methadone is 1.92 (ED_{50} = 1.30) and that of pethidine 0.32 (ED_{50} = 7.7).

Figures 13, 14 and 15 show that it was also possible to evaluate, and establish a quantitative difference between the duration of effect of the increasing doses of each drug by this method.

In the case of *morphine*, the slope of the rate-of-action curve increased proportionately to the dose, the peak effect being reached, whatever the dose, towards the second hour after

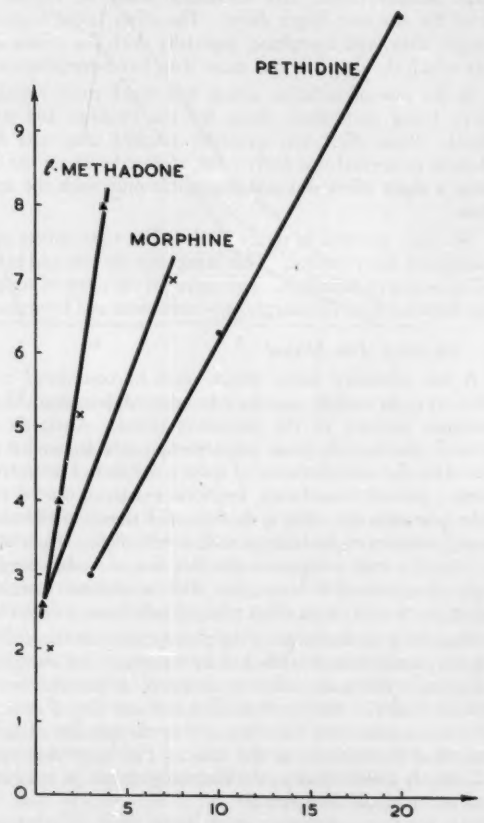


FIGURE 19

Effect/dose ratio for morphine, pethidine and l-methadone

x axis : doses in mgr/kg

y axis : rise in the threshold of response in excitation units (Ex.U.)

TABLE III

Drug →	*Morphine*			*Pethidine*			*Methadone*		
Dose ratio → *(mg/kg)*	*2.5 to 0.8*	*7.5 to 2.5*	*7.5 to 0.8*	10 to 3	*20 to 10*	20 to 3	*1.25 to 0.4*	*3.75 to 1.25*	*3.75 to 0.4*
"t" value	1.97	1.42	2.90	3.22	3.85	6.65	1.59	2.85	4.26
P	>0.05	>0.05	<0.05	<0.05	<0.01	<0.01	>0.05	<0.05	<0.01

administration. Return to normal was also gradual and varied from the third hour for a weak dose (0.8 mg/kg), the fourth hour for a medium dose (2.5 mg/kg), to the fifth hour for a strong dose (7.5 mg/kg).

In the case of *methadone* there was also a progressive slope becoming steeper and steeper with the increase in dose, peak effect being reached, as with morphine, about two hours after administration, and remaining more or less at that level for the two larger doses. The effect lasted somewhat longer than with morphine, especially with the strong dose, for which the duration was more than five hours (figure 15).

In the case of *pethidine*, action was much more rapid, the slope being particularly sharp for the medium and strong doses. Peak effect was generally reached after one hour. Return to normal was fairly swift, so that by about the third hour a slight effect was still discernible only with the strong dose.

We have grouped in figure 20 the effect time curves of the maximum doses studied. This brings out the rate and brevity of the action of pethidine. The curve for the onset of analgesia has the same shape for morphine, *l*-methadone and levorphanol.

C. *Electivity of the Method*

A last reliability factor which must be considered is the *electivity of the method*—i.e., the possibility of detecting the true analgesic potency of the substances tested. Analgesia is a general pharmacodynamic phenomenon which can be produced by the administration of quite a number of neurotropic drugs : general anaesthetics, hypnotic sedatives, central muscular relaxants, etc. But in the case of all these " pseudo-analgesic " substances, analgesia is only a side-effect. The characteristic of a true analgesic is the fact that the suppression of pain constitutes an isolated effect with an adequate margin of safety in relation to its other pharmacodynamic actions (163).

Thus the post-discharge of the polysynaptic circuits following the painful stimuli is blocked by morphine and morphinomimetics. The same effect is obtained, as pointed out by Wikler et al. (72, 161) with barbiturates and central muscular relaxants (myanesine), but whereas with the narcotic analgesics this effect is selective, in the case of the other depressants it is merely a side-effect to the inhibiting action on consciousness and postural integration.*

This fundamental difference is also evident in the results obtained in the parallel study of narcotic analgesics and the central depressants. On the one hand we observed no hypnotic sedative effect with the analgesic doses of narcotic analgesics in guinea-pigs. The animals were brisk and wide awake. The quality of the P.R.T. was unchanged, and the movement was as clear-cut and swift as on grading. The margin of safety for toxic side-effects was $I_{50} = 6.8$. There is thus every reason to believe that the increase in the threshold value is really due to analgesic action.

With the central depressants which were studied, on the other hand, analgesic action could be detected only with anaesthetizing doses. Only then could a large elevation of the threshold of response be observed. This side-effect analgesia was of short duration with ether (figure 17), but several hours long with thiopental (figure 18). This cross-effect is also evidenced by a change in the form of the P.R.T. The movement is sluggish, and even completely absent in certain animals. With non-hypnotic doses of thiopental and ether we obtained little or no analgesic effect (figures 17 and 18).

When discussing this question one must bear in mind that the responses are somatic defensive reactions and that a certain muscular relaxation induced by strong doses of anaesthetics may distort the analgesic action.

It follows that, on the whole, use of the electro-dental method on guinea-pigs revealed an undoubted electivity in analgesic action in the case of morphine and morphinomimetics.

With dogs, it seems that analgesic doses already tended to have a slight dormitive action. The doses of morphine studied by Koll & Reffert (91) were 1.25 and 3.75 mg/kg, while those of Fleisch & Dolivo (41) ran from 0.5 to 3.75 mg/kg. For dogs, however, the depressive dose of morphine is 2 mg/kg. Hatcher & Eggleston (59), in a series of systematic studies, even put it at less than 1 mg/kg.

With the electro-dental method used on rabbits, the analgesic doses lie between 2.5 and 5 mg/kg (36). Hypnotic effect is already present with 2–3 mg/kg doses (36, 131). Respiratory depression is already evident at 0.15 mg/kg (37), and the diminution in the minute volume reaches 50 per cent with a 3 mg/kg dose (177). Such effects alter the experimental conditions, and the analgesic effect can be put down at least in part to hypnosis.

II. VALIDITY OF THE METHOD

1. *Comparison with Results of Animal Experimentation*

After studying the reliability of the method from the physiological and pharmacological standpoints, we must now test the validity of the results by comparing them with other data obtained in experiments with animals and in man. Any parallel with the whole gamut of mechanical, chemical

* A useful complement to Wikler's work is provided by that of French, Verzeano & Magoun 1953 (42, 43) on the neurophysiological basis of the action of ether and pentobarbital.

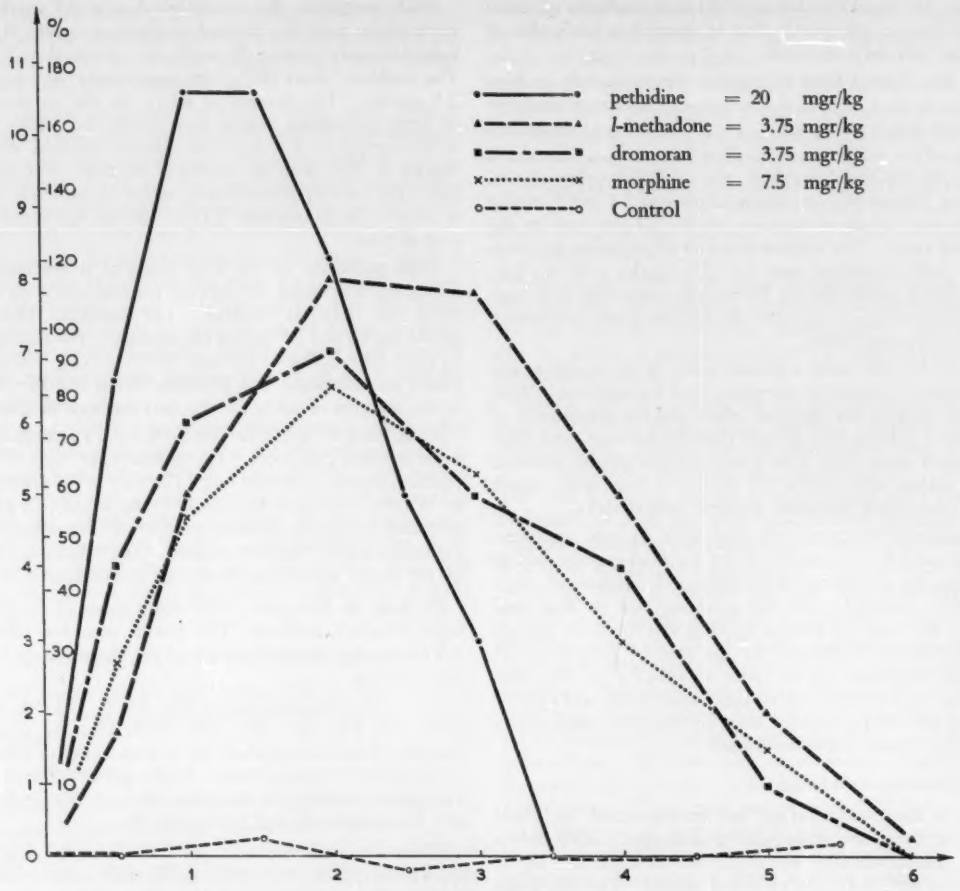


FIGURE 20

Analgesic action of morphine, pethidine, l-methadone and levorphanol. Curve : effect/time of maximum doses studied
x axis : time in hours
y axis : rise in the threshold of response in Excitation Units (Ex.U.) and in %

and electro-cutaneous methods used in *animal experiments* being outside the scope of this work, our comparison will be confined to the electro-dental and thermo-cutaneous methods.

A. *Electro-dental Methods*

The analgesic effect of morphine and the morphinomimetics, as recorded by the electro-dental method, varies according to the author.

On dogs, Koll & Reffert (91) studied the action of morphine and some semi-synthetic derivatives. With four dogs, the standard doses of morphine, 1.25 and 3.75 mg/kg, led to heavy increases in the threshold value, of 85 per cent and 171 per cent respectively. Two dogs showed a tendency to sleepiness which the author attributes not to the narcotic effect of the morphine, but, after checking with codeine, to a sluggishness of the motor system brought about by narcotic analgesics.

Soehring notes a very marked elevation of the threshold of response with 1 mg/kg of morphine and a limited rise in the Z syndrome value. Is it possible to strike a parallel with the results obtained in guinea-pigs where narcotic analgesics also show a distinctly stronger action on the Pain Reaction Test than on the R_0 response ? Whether this is a pharmacological proof that R_0, like Soehring's (144) Z syndrome is not a reaction to pain, but the earliest stage in the motor effect induced by excitation, is a difficult question to answer. The data in medical literature regarding the action of morphine on simple reflexes are still contradictory. Mazoué, 1927 (111), found that morphine reduced the reflex excitability of the marrow, while Luckard & Johnson, 1928 (109), obtained inhibition of the patellar reflex in cats. Houde & Wickler (72) and Wickler (161), on the other hand, have recently reported that with spinal dogs morphine does not depress the monosynaptic (patellar) reflex or the bi-synaptic reflex (ipsi-lateral extensor reflex).

Recently Boréus, 1955 (17), also made a comparative study of the analgesic effect on dogs of morphine and three synthetic morphinomimetics : *l*-methadone, pethidine and ketobe-

midone. He found that, for equal doses, the intensity of action of *l*-methadone was double that of morphine, while that of pethidine was only one-sixth.

The data derived from stimulating the tooth pulp in dogs show the method to be highly sensitive. Significant analgesic effects can be recorded with a dose of 1 mg/kg of morphine.

Study of the analgesic action of narcotic analgesics in rabbits by the electro-dental method gave the following results. Fleisch & Dolivo (41), on administration of 2.5 and 5 mg/kg of morphine, obtained increases of 70–200 per cent in the threshold value. The relative potency of pethidine by comparison with morphine was 1/4 (2.5 mg/kg : 10 mg/kg). The analgesic action did not seem to be pure with such high doses, there being, according to the authors, some interference from the hypnotic effect.

Yim et al. (178) made a parallel study of the analgesic and the bradypneic action of morphine and levorphanol. With the doses studied, the analgesic effect and the diminution of the minute volume was greater than 50 per cent, and their effect lasted more than 3 to 4 hours. The results obtained by this author with rabbits are similar to ours with respect to the levorphanol morphine analgesic action ratio.

In short, the results we obtained with narcotic analgesics in guinea-pigs by the electro-dental method may be said to tally from the standpoint of intensity and duration of effect with those recorded with the same method in dogs and rabbits. The order of relative potency was the same, though there was some discrepancy in the case of pethidine, the guinea-pig appearing to be twice as sensitive to this drug as the dog or the rabbit. A narcotic effect (dogs and rabbits) and other side-effects (rabbits) may, on the other hand, interfere with analgesia at the doses used.

B. *Thermo-cutaneous Methods*

As far as *thermo-cutaneous methods* are concerned, we dwell mainly on the work done with guinea-pigs. Although a different painful stimulus was used, this work had one factor in common with ours—the choice of animal. The thresholds of sensitivity to drugs and their metabolism were thus constants.

Hildebrandt, 1934 (69), using the nociceptive direct-heat method, recorded a minimum analgesic action with a dose of 5 mg/kg of morphine, the duration of effect being 60 minutes. It was not until the dose was increased to 20 mg/kg that a 50 per cent increase in the threshold value (40° to 60°) was obtained with a duration of effect of about six hours. Thus, with the same animal, the doses were 6–8 times as great as with dental stimulation.

We deal more fully with the radiant heat methods introduced by Hardy-Wolff-Goodell (54), and d'Amour Smith (28), as they now constitute the routine laboratory procedure in algesimetry. Apparently this popularity is primarily due to the fact that the heat ray, as pointed out by Jacob (82), is the most selective of the painful stimuli in the mixed sensorial field. Furthermore, with this method parallel quantitative measurements can be made in man and animals.

Since Winder (166, 167, 168), in a thorough statistical study, laid the algesimetric basis for using this method in guinea-pigs, observing the analgesic action and toxicity of two narcotic analgesics, morphine and pethidine, it might be of interest to compare their results with ours with respect to absolute analgesic effect and the margin of safety for toxic side-effects.

With morphine, the minimum dose is 3.2 mg/kg, intraperitoneally with the thermal method, as against 0.8 mg/kg subcutaneously (figure 9) with the electro-dental method. The analgesic doses (ED_{50}) are respectively 20.3 mg/kg and 2.5 mg/kg. The margin of safety for the commencement of toxic side-effects which may distort the results is I_{50} = 32/20 mg/kg = 1.6 with the thermal method and 32/2.5 mg/kg = 12.8 with the electrical method. The margin of safety for lethality is less important for algesimetry, especially as lethality in guinea-pigs (LD = 390 mg/kg) is much lower than in man.

With pethidine, 20 per cent analgesia is obtained with a 26 mg/kg dose using the thermal method and with 3 mg/kg using the electrical method. The analgesic doses (ED_{50}) are 80 mg/kg and 7.7 mg/kg respectively. The corresponding I_{50} are 53/80 mg/kg = 0.6 and 53/7.7 = 6.8. The Index 50 values for morphine and pethidine bring out the difference in the margins of safety for the two methods in guinea-pigs.

It can thus be seen that despite Winder's remarkably strict experimental conditions, the radiant-heat method is less practical than the electrical one. The very heavy doses required in Winder's work, as Bianchi & Franceschini (13) point out, may also be due to administration by the less effective intraperitoneal route. Narcotic analgesics thus injected are absorbed by the portal system and destroyed in the liver (129, 151).

De Jong & Knoppers, 1952 (86), obtained similar results with Winder's method. The lowest dose for which they recorded a significant elevation of the threshold ($p = 0.002$) was 5 mg/kg.

The results obtained in rats (16, 22, 51, 53, 73, 84, 119, 128*b*, 143, 156, 158 169, 169*a*, 169*b*, 170) by radiant and hot-plate heat methods show the average analgesic dose (ED_{50}) to be about 4 mg/kg (range : 2–10 mg/kg). The depressive dose in rats is estimated, according to the author, at 4.5 mg/kg (37), 5 mg/kg (131) and 2.1 mg/kg (7).

When the same methods are used with mice (38, 52, 53, 63, 81, 94, 98, 99, 117, 176), the average value of the analgesic doses is very near to that for rats : 3.8 mg/kg (range : 1.6–8.6 mg/kg). But with doses of less than 1 mg/kg, mice already show a psychoneurotic state which might interfere with the analgesic action. Radouco-Thomas C., Radouco-Thomas S. & Nosal Gl. (126) have recently studied the relation between the analgesia and the psychomotor state induced by morphine and the synthetic morphinomimetics in animal experiments (cats and mice).

It emerges from all these comparisons that with radiant or hot-plate heat methods, whatever the animal (guinea-pig, rat or mouse), the side-effects, depression or hyper-excitability of the central nervous system, interfere with the analgesic action. Other drawbacks to these methods are :

Stimulation of two types of receptor;

Difficulty of obtaining thermal stability at the level of the pain receptors, which may alter the value of the actual stimulus and conceal the true degree of analgesia (50, 120, 164, 170);

Biological reactions which, though involving long supraspinal circuits (72, 75), have not the complexity of the reactions obtained by the tooth pulp stimulation. Narcotic analgesics, however, act selectively on the complex reactions (82) mainly of a preponderant type.

On the other hand, the way in which the potential stimulus is measured (the caloric energy applied), the steadiness of the

threshold value (in terms of heat or reaction time), the reproducibility and the constancy of the biological response generally elicited from small laboratory animals make this method suitable for mass experiments.

Hence thermal methods, especially those using radiant heat, have become the most generally used in algesimetry.

Comparison of the algesimetric results obtained in experiments with animals by the two main groups of methods—the thermocutaneous and the electro-dental—seems, however, to suggest that the latter are to be somewhat preferred.

We do not thereby wish to imply that one group of methods should be chosen rather than the other for the study of analgesics. We merely wish to bring out the fact that when guinea-pigs are used for the stimulation of dental receptors, the method ceases to be a luxury suited only for pure research, and lends itself to routine laboratory work on a par with the other techniques.

Pathological pain is a subjective phenomenon which takes on several forms (superficial, deep and visceral pain) and is very complex in origin. The value of the experimental methods, though real, is limited, for they provide information only on the physiological reactions (somato-vegetativo-humoral) to a painful stimulus. This deficiency has to be remedied by using stimuli of the most varied kinds and sources. A whole battery of methods is therefore essential for analgesimetric studies.

2. *Comparison with Results obtained in Man*

We will now compare our results with the data obtained in man for induced (experimental) pain and natural (pathological) pain.

A. *Experimental Algesimetry*

The main varieties of experimental (superficial, deep and visceral) pain will be considered. Schaumacher et al., 1940 (139), Holland & Gross, 1946 (70), Miller, 1948 (114) and Christensen & Gross, 1948 (27), obtained very favourable results by the radiant-heat method and administration of morphine and morphinomimetics. Christensen & Gross, 1948 (27), using the Hardy-Wolff-Goodell method (55), found methadone administered subcutaneously three times as potent as morphine and several times as potent as pethidine. The peak action effect comes about 90 minutes after administration, and duration of effect for the three analgesics ranges from 3 to 6 hours. The radiant-heat method, regarded as the best for studying *superficial pain*, has not, however, always given concordant results. The work of Dodds, 1945 (32), Thorp, 1946 (154), Denton & Beecher, 1949 (30) and Kuhn & Bromiley, 1951 (96), points to the uncertainty of the data in man.

In the study of *deep pain*, results have been obtained by the ischemic contraction of the muscle and by the electro-dental method. The method of ischemic contractions of muscle was introduced by Lewis et. al. (101, 102), to study pain and adopted by Harrison & Bigelow, 1943 (58), to evaluate analgesia. Hewer et al. 1948 (67), 1949 (68), made a statistical study of the effect of psychogenic factors and analgesic medication on this type of deep pain. They observed that, even with the very great variations caused by the former, morphine, pethidine and methadone had significant action ($P = 0.02$). Measurements show that 30 mg/kg of *l*-methadone have a similar analgesic action to 32 mg/kg of morphine and greater than that induced by 150 mg/kg of pethidine.

Let us now consider the algesimetric results obtained by the electro-dental method in man, the forerunner of that which we used with animals.

Heinroth, 1926 (61), obtained a marked elevation of the threshold (90 per cent) with 10 mg of morphine administered subcutaneously. The effect reached its acme after about one hour and lasted about five hours.

Goetzl, 1943 (50), found a marked elevation of the threshold with narcotic analgesics, which was not confirmed under similar technical and pharmacological conditions by Harris & Blockus, 1952 (57).

Pfeiffer et al., 1948 (120), using this method, made a distinction between the threshold of perception (minimum pain) and the threshold of response (severe pain). They observe that the action of narcotic analgesics is much stronger at the threshold of response.

Boréus, 1955 (17), evaluating analgesic action on the central incisors by the electro-dental method, obtained for doses equivalent to 10 mg, the strongest effect with morphine ($P = 0.01$-0.001) followed by *l*-methadone ($P = 0.02$-0.01). The peak action of morphine was towards the second hour. Pethidine, in a 65 mg dose, produced no significant effect. The action was recorded on the first distinct painful sensation. It is to be noted that a pronounced decrease in the threshold values of the controls was recorded during the experiment.

Visceral algesimetry is still in its infancy. Little work has been done on it (21, 24, 112, 171). Gaensler et al., 1948 (46), 1950 (47), 1951 (48), determined the analgesic action of morphine (10 mg) and the morphinomimetics pethidine (100 mg), *l*-methadone (7.5 mg) and levorphanol (3 mg), on the threshold of visceral pain induced by distension of the biliary ducts. With the doses administered, morphine proved most active (56 per cent), followed by dromoran (42 per cent), *l*-methadone (33 per cent) and pethidine (23 per cent). The acme was reached after about 30 minutes with morphine and meperidine (pethidine) and about one hour with the other substances. Duration of effect, in rising order of magnitude, was three hours for pethidine, five for morphine and six for *l*-methadone.

Apart from the difference with regard to the potency of morphine and its precocious peak action, a fairly good concordance is to be observed between the algesimetric data in guinea-pigs and the action of analgesics on visceral pain in man. In the same work, the author brings out the strong influence of psychological and physiological components on analgesic activity by studying simultaneously the threshold of response when asleep and that of perception when awake.

This brief review of the algesimetric methods used to induce experimental pain in man shows that there are many irregularities and even contradictions in the results obtained. This defect is probably due to too great variability in the biophysical parameters (insufficient regularity in the actual stimulating force) and the psychological parameters.

With regard to the former, Björn (14) and Harris (57) have succeeded, with electro-dental methods, in obtaining relatively high stability of the actual stimulating force. With thermal methods the problem seems more difficult to solve owing to the thermal instability of the stimulated place itself.

Whatever the method used, however, there still remain the psychological variables (48, 58, 68, 175) which are a serious handicap in experiments with man. When pain is induced in man, the threshold of psychic, anticipative and associative

reactivity, unlike that of perception, shows great variability (24, 48, 57, 70, 106, 145, 171, 172, 173) due more to the affective tonus of the subject than the strength of the stimulus. The use of selected and trained subjects, the administration of placebos and the device of " dry runs " are only partial remedies. The objective study of analgesia seems to call for subjects with a constant threshold of response, in other words subjects in which a state of analgesia has already been induced.

In experiments with animals, the somatic reactions " visible and measurable evidence of pain " are less varied than the somatic reactions of man. Yet narcotic analgesics seem to act less on the perception factor than on the psychic and somatic reactions. The great variation in the thresholds of psychic response in man and the steadiness of the somatic response thresholds in animals explain the inconsistent results in human experiments and the great consistency of those obtained in experiments with animals—which have, furthermore, the advantage of great numbers. The results of animal experiments, paradoxical as that may seem, may therefore be regarded as coming closer to clinical analgesic action than those of human experiments, always assuming that the somatic reactions of the animal are accepted as genuine evidence of pain.

B. *Pathological Pain*

Let us now consider the data of human pathology which give final sanction to any experimental result. In Table IV, the analgesic action of the three substances studied is expressed in terms of effective doses and their relative potency by comparison with morphine. The experimental data are the ED_{50} while the clinical data are the traditionally accepted average values, regardless of individual factors such as the intensity, duration (acute or chronic) or seat of pain (superficial, deep or visceral).

TABLE IV

	Man		*Guinea-pigs*	
Narcotic analgesics	*ED (mg. total)*	*Relative potency*	*ED_{50} (mg./kg.)*	*Relative potency*
Morphine	5–15	1	2.5	1.0
l-Methadone	5	2	1.3	1.9
Pethidine	50–150	0.1	7.7	0.3

It will thus be seen that the analgesic intensity met with in clinical practice is similar to that obtained in guinea-pigs. This similarity applies both to the sequence and the order of magnitude in relation to morphine. Some qualification must be made in the case of pethidine, to which the guinea-pig, unlike the dog (17), is more sensitive than man.

As for the duration of effect, the progressive series, pethidine, morphine, *l*-methadone of clinical practice was also encountered in our experiments. The maximum doses studied have a duration of effect of the same order of magnitude as that of average clinical doses.

III. Efficiency of the Method

After the reliability and the validity of the electro-dental method in guinea-pigs, there remains a last criterion to consider—that of efficiency.

With dogs, the electro-dental method, although probably the best algesimetric method, has been replaced as a routine laboratory procedure by the thermo-cutaneous methods. " The disadvantages of the tooth pulp algesimetry as used in animals is that it is too time-consuming, cumbersome and expensive. Consequently, it cannot be recommended as a routine laboratory procedure " (17).

The method we advocate avoids these disadvantages. The choice of a small laboratory animal, inexpensive and easy to handle, together with the use of a system of electrodes easily inserted and of simple design makes it possible for a series of experiments to be performed by a single experimenter.

The use of a large number of animals makes it possible to achieve greater accuracy of results (systematic error $\bar{x} = \frac{\sigma}{\sqrt{N}}$), to meet the growing demands of chemical synthesis, to avoid the acute tolerance phenomena observed in experiments with narcotic analgesics (38b, 38c, 51, 95).

Electro-dental stimulation in guinea-pigs, therefore, seems to meet all the requirements, and may be regarded as a routine algesimetric procedure. There is, however, another bridge to be crossed before final judgment can be passed. We must wait until the results of the method will be reproduced and confirmed by other research workers.

Summary and Conclusions

The systematic reproduction of pain and evaluation of the analgesic potency of morphine and some synthetic morphinomimetics; pethidine, *l*-methadone and levorphanol, have been effected in animal experiments (with guinea-pigs) by a new electro-dental algesimetric method.

1. *The reliability of the method* has been checked from the physiological and pharmacological standpoints.

2. A series of " somatic physiological responses " R_0, R_1, R_2, has been induced by electrical stimulation of the tooth pain receptors of guinea-pigs. Response R_1 (upward thrust of the head) was selected as the Pain Reaction Test (P.R.T. = R_1). Neuro-physiological data make it possible to regard this as true evidence of a sensation of pain.

3. The P.R.T. is capable of qualitative and quantitative definition. It is easy to identify, constant and of reproducible form. Quantitatively, the P.R.T. is defined by the threshold voltage. The stability of the potential stimulus and, what is more, of the actual stimulus (stimulus-receptor complex) has been confirmed by the almost complete constancy of the threshold of response during the same experiment.

4. The analgesic potency of morphine and the three synthetic morphinomimetics, pethidine, *l*-methadone and levorphanol, has been evaluated from the increase in the threshold of the P.R.T. and expressed in the dose/effect/time ratio.

5. The sensitivity of the method has been proved by the weakness of the doses required to produce minimal analgesia.

6. The selectivity of the method has been revealed in two respects : intensity and duration of effect.

In descending order of potency we have : *l*-methadone, levorphanol, morphine and pethidine.

The relative potency, by comparison with morphine (ED_{50} = 2.5) taken as one, is 1.9 for methadone (ED_{50} = 1.3) and 0.32 for pethidine (ED_{50} = 7.7).

The peak effect is reached after one hour with pethidine and after two hours with the three other drugs.

Duration of effect varies with the dose from about 2–3 hours for pethidine to 4–6 hours for morphine, levorphanol and *l*-methadone.

7. The electivity of the method when used with narcotic analgesics has been demonstrated by a double check : the inducing of analgesia as an isolated effect in the case of morphine and the morphinomimetics (wide margin of safety for toxic side effects and lethality); the inducing of analgesia as a side-effect of the depressants studied—i.e., ether and thiopental (cross-effect of general anaesthesia and analgesic action).

8. *The validity of the method* has been demonstrated by a comparison with the electro-dental and thermo-cutaneous methods used in animal experiments. Its validity has been finally established by comparison with the results obtained in man with induced pain (superficial, deep and visceral) and with spontaneous (pathological) pain.

9. *The efficiency of the method* is disputed. The use of a laboratory animal lending itself to statistical study, of simple apparatus and simple laboratory technique makes it possible to regard the electro-dental stimulation of guinea-pigs as a routine algesimetric procedure for studying new analgesics.

Aeknowledgements

The authors wish to thank Dr. A. Linder, Professor of Statistics in Geneva for his valuable advice with the biometric analysis of the results we obtained.

We wish also to express our deep appreciation to Dr. E. Le Breton, Professor of Physiology at the University of Paris, who made it possible for us to do the present work, and to Dr. Ed. Frommel, Professor of Pharmacodynamics in Geneva, who permitted us to conduct part of our experiments in his laboratory. We wish, too, to convey our thanks to Mr. Ph. Gold, chemical Engineer, and to Mr. L. Strassberger, Technical Assistant at the Institute of Therapeutics of Geneva, for their valuable collaboration.

We wish to convey our thanks also to Dr. J. Scherrer, *Professeur agrégé* at the Faculty of Medicine at the University of Paris, for his kindness in placing at our disposal before their publication the manuscripts of the Symposium on Pain.

BIBLIOGRAPHY

1. Alajouanine : *La douleur et les douleurs*, Masson, Paris, 1956.
2. Amsler, C. : Sind Schrei und Abwehrbewegungen nach Schmerzreizen beim normalen Tier Zeichen empfundenen Schmerzes oder nur reflektorische Erscheinungen, *Arch. f. exp. Pathol. u. Pharmakol.*, 1921, *90*, pp. 257–276.
3. Andrews, H. L. : Cortical Effects of Demerol, *J. Pharmacol.*, 1942, *76*, pp. 89–94.
3*b*. Andrews, H. L. : Skin Resistance Changes and Measurements of Pain Threshold, *J. Clin. Invest.*, 1943, *22*, pp. 517–520.
4. Andrews, H. L. : The Effects of Opiates on the Pain Thresholds in Post-addicts, *J. Clin. Invest.*, 1943, *22*, pp. 511–516.
5. Andrews, H. L. : Changes in the Electroencephalogramm during a Cycle of Morphine Addiction, *Psychosom. Med.*, 1943 *5*, pp. 143–147.
6. Auluck, F. C. & Kothari, D. S. : Radiation and Sensation of Pain, *Nature*, 1949, *164*, p. 923.
7. Barlow, O. W. : The tranquilizing Potency of Morphine, Pantopon, Codeine, Papaverine and Narcotine, *J. A. M. A.*, 1932, *99*, p. 986.
8. Baud, C. A. & Held, A. J. : Innervation de l'organe dentaire, Reports and Papers, *XIIIth Congress Internat.*, Assoc. les Paradont, Geneva, 1953, p. 180.
9. Beckett, H. H. : Analgesics — A general Survey, *J. Pharm. & Pharmacol.*, 1952, *4*, pp. 425–477.
10. Beecher, H. K. : Analgesic Power and the Question of Acute Tolerance to Narcotics in Man, *J. Pharmacol.*, 1953, *108*, 2, pp. 158–167.
11. Behrens, B. & Karber, G. : Quelle est la façon la plus opportune d'arranger des séries d'expériences pour un enregistrement biologique? *Arch. f. exp. Pathol. u. Pharmacol.*, 1935, *177*, p. 379.
12. Berkelbach van der Sprenkel, H. : Treten peridontale Nervenfasern in das Dentin über? *Ztschr. mikr.-anat. Forsch.*, 1934, *36*, p. 509.
13. Bianchi, C. & Franceschini, J. : Experimental Observations on Haffner's Method for Testing Analgesic Drugs, *Brit. J. Pharmacol.*, 1954, 9, pp. 280–284.
14. Björn, H. : *Swed. Dent. J.*, 1946, 39, suppl., - quoted by Harris & Blockus (57).
15. Bonica, J. J. : *The Management of Pain*, Lea & Febiger, Philadelphia, 1953.
16. Bonnycastle, D. D. & Leonard, C. S. : An Estimation of the Activity of Analgetic Materials, *J. Pharmacol.*, 1950, *100*, pp. 141–145.
17. Boréus, L. O. & Sandberg, F. : A Comparative Study of Different Algesimetric Methods in Animals and Man, *Acta pharmacol. & toxicol.*, 1955, *11*, pp. 198–214.
18. Braenden, O. J., Eddy, N. B. & Halbach, H. : Synthetic Substances with Morphine-like Effect — Relationship between Chemical Structure and Analgesic Action, *Bull. Wld Hlth Org.*, 1954, *10*, pp. 1003–1038.
19. Brashear, A. D. : The Innervation of the Teeth, *J. Comp. Neurol.*, 1936, *64*, p. 169.
20. Brock, N. & Gex, F. : Die Bestimmung des Therapeutischen Breite von Arzneimitteln, *Arzneimittelforschung*, 1951, *2*, p. 63.
21. Butsch, W. L., McGowan, J. M. & Walters, W. W. : Clinical Studies on the Influence of certain Drugs in Relation to Biliary Pain and to the Variations in Intrabiliary Pressure, *Surg., Gynec. & Obst.*, 1936, *63*, pp. 451–456.
22. Cahen, R., Epstein, H. & Krementz, C. : The Evaluation of the Analgesic Action of Methadone Isomers and other Analgesics by a new rapid Screening Method, *J. Pharmacol.*, 1948, *94*, p. 328.
23. Cahen, R. & Wikler, A. : Effects of Morphine on Cortical Electrical Activity of the Rat, *Yale J. Biol. & Med.*, 1944, *16*, pp. 240–243.
24. Chapman, W. P., Herrera, R. & Jones, C. M. : A Comparison of Pain produced experimentally in Lower Oesophagus, Common Bile Duct and Upper Small Intestine with Pain experienced by Patients with Diseases of Biliary Tract and Pancreas, *Surg. Gynec. & Obst.*, 1949, *89*, p. 573.
25. Cook, H. F. : The Pain Threshold for Microwave and Infra-red Radiations, *J. Physiol.*, 1952, *118*, pp. 1–11.
26. Christensen, K. : Sympathetic Nerve Fibres in the Alveolar Nerves and Nerves of the Dental Pulp, *J. D. Res.*, 1940, *19*, p. 227.
27. Christensen, E. M. & Cross, E. G. : Analgesic Effects in Human Subjects of Morphine, Meperidine and Methadone, *J. A. M. A.*, 1948, 137, pp. 594–599.
28. D'Amour, F. E. & Smith, D. L. : A Method for Determining Loss of Pain Sensation, *J. Pharmacol.*, 1941, *72*, pp. 74–79.
29. Davies, D. : Clinical Testing of Analgesics, *Acta pharmacol. & toxicol.*, 1954, *10*, pp. 113–116.
30. Denton, J. E. & Beecher, M. D. : New Analgesics. — I. Methods in the Clinical Evaluation of New Analgesics, *J. A. M. A.*, 1949, *141*, pp. 1051-1057.
31. Dependorf, L. : Beiträge zur Kenntnis der Innervierung der menschlichen Zahnpulpa und des Dentins, *Deutsche Monatschr. Zahnh.*, 1913, *31*, p. 689.
32. Dodds, E. C., Lawson, W., Simpson, S. A. & Williams, P. C. : Testing Diphenyl-ethylamine Compounds for Analgesic Action, *J. Physiol.*, 1945, *104*, pp. 47–51.
33. Domenjoz, R. : Some Pharmacological Aspects of Phenylbutazone (Butazolidine), *Intern. Record of Medicine*, 1952, *165*, 9, pp. 467–472.
34. Domenjoz, R. : Pharmacological Evaluation of Anti-Inflammatory Agents, a Contribution of Phenylbutazone (Butazolidine), *Ann. Univ. Saraviensis*, 1953, *4*, pp. 317–344.
35. Dorpat, T. L. : *Mechanisms of Muscle Pain*, Med. Thesis, Washington, 1952.
36. Dott, D. B. & Stockman, R. : Pharmacology of Morphine and its Derivatives, *Proc. Roy. Soc.*, 1890, *17*, p. 321.
37. Eddy, N. : Pharmacology of Metopon and other New Analgesic Opium Derivatives, *Ann. N. Y. Acad. Sc.*, 1948, *51*, 1, pp. 51–58.

38. Eddy, N., Touchberry, C. & Liebermann, J. : Synthetic Analgesics — I. Methadone Isomers and Derivatives, *J. Pharmacol.*, 1950, *98*, pp. 121–137.

38*b*. Ercoli, N. & Lewis, M. N. : Studies on Analgesics — I. The Time-Action Curves of Morphine, Codeine, Dilaudid and Demerol by Various Methods of Administration, *J. Pharmacol.*, 1945, *84*, pp. 301–307.

38*c*. Fichtenberger, D. : Study of Experimental Habituation to Morphine, *Bull. of Narcotics*, 1951, III, 3 and 4, pp. 20–44, 17–52.

39. Finney, M. A. : *Probit Analysis, A Statistical Treatment of the Sigmoid Response Curve*, University Press, Cambridge, 1947.

40. Fisher, R. A. : *Statistical Methods for Research Workers*, Oliver & Boyds, London, 1936.

41. Fleisch, A. & Dolivo, M. : Auswertung der Analgetica im Tierversuch, *Helv. Physiol. Acta*, 1953, *11*, pp. 305–322.

42. French, J. D., Verzeano, M. & Magoun, H. W. : An Extralemniscal Sensory System in the brain, *A. M. A. Arch. Neurol. & Psychiatr.*, 1953, *69*, pp. 505–518.

43. French, J. D., Verzano, M. & Magoun, H. W. : A Neural Basis of the Anaesthetic State, *A.M.A. Arch. Neurol. & Psychiat.*, 1953, *69*, pp. 519–529.

44. Freund, H. : Zur Pharmakologie der Schmerzstillung, *Arch. f. exp. Pathol. u. Pharmakol.*, 1936, *180*, p. 209.

45. Frommel, Ed. et al. : Le N-acétyl p-aminophénol, *Praxis*, 1953, 46, pp. 968–972.

46. Gaensler, E. A., McGowan, J. M. & Henderson, F. F. : A comparative Study of the Action of Demerol and Opium Alkaloids in Relation to Biliary Spasm, *Surgery*, 1948, *23*, p. 211.

47. Gaensler, E. A., McGowan, J. M. & Henderson, F. F. : Biliary and Small Intestinal Spasm due to Synthetic Analgesics : Methadone and Methyldihydromorphine (Metopon), *Gastroenterology*, 1950, *16*, p. 484.

48. Gaensler E. A. : Quantitative Determination of Visceral Pain Threshold *J. Clin. Invest.*, 1951, *30*, pp. 406–422.

49. Gangloff, H. & Monnier, M. : Demonstration de l'impact des analgésiques morphiniques et d'une substance antimorphinique sur le cortex et le sub-cortex du lapin, *Helv. Physiol. Acta*, 1955, *13*, *4*, pp. C. 46–47.

50. Goetzl, F. R., Burrill, D. Y. & Ivy, A. C. : A Critical Analysis of Algesimetric Methods with Suggestions for a Useful Procedure, *Bull. Quart. Northw. Univ. School* (Chicago), 1943, *17*, pp. 280–291.

51. Green, A. F., Young, P. A. & Godfrey, E. J. : A Comparison of Heat and Pressure Analgesiometric Methods in Rats, *Brit. J. Pharmacol.*, 1951, *6*, pp. 572–587.

52. Grewald, S. : A Method for Testing Analgesics in Mice, *Brit. J. Pharmacol.*, 1952, *7*, pp. 433–437.

53. Haas, H., Hohagen, E. & Kolimannsperger, G. : Vergleichende Untersuchungen mit Analgetics, *Arzneimittelforschung*, 1953, *3*, pp. 238–247.

54. Hardy, J. A., Wolff, H. G. & Goodell, H. : Studies on Pain : A New Method for Measuring Pain Threshold : Observation on Spatial Summation of Pain, *J. Clin. Invest.*, 1940, *19*, pp. 649.

55. Hardy, J. A., Wolff, H. G. & Goodell, H. : The Pain Threshold in Man, *Amer. J. Psychiatr.*, 1943, *99*, p. 744.

56. Hardy, J. A., Wolff, H. G. & Goddell, H. : *Pain Sensations and Reactions*, Williams & Wilkins Co., Baltimore, 1953.

57. Harris, S. C. & Blockus, L. E. : The Reliability of Validity of Tooth Pulp Algesimetry, *J. Pharmacol.*, 1952, *104*, 2, pp. 135–148.

58. Harrison, I. B. & Bigelow, N. H. : Quantitative Studies of Visceral Pain produced by the Contraction of Ischemic Muscle, *A. Research Nerv. & Ment. Disc. Proc.*, 1943, *23*, p. 154.

59. Hatcher, R. A. & Eggleston, C. : Studies on the Absorption of Drugs, *J.A.M.A.*, 1914, *63*, p. 469.

60. Hazard, R. Pharmacodynamie des Analgésiques — in *Douleur*, Alajouanine, Paris, 1956 (in the press).

61. Heinroth, H. : Ueber die Wirkung verschiedener Arzneimittel auf die Schmerzempfindlichkeit der Zahnpulpa, *Arch. f. exp. Pathol. u. Pharmakol.*, 1926, *116*, pp. 245–260.

62. Held, A. J. & Baud, C. A. : The Innervation of the Dental Organ, *Oral Surgery Medicine & Oral Pathology*, 1955, *8*, 12, pp. 1262–1269.

63. Herr, F. & Porszasz, J. : *Arch. f. exp. Pathol. u. Pharmakol.*, 1950, *210*, p. 289.

64. Hess, W. R. : Das Zwischenhirn als Koordination Organ, *Helv. Physiol. Acta*, 1943, 1, pp. 549–565.

65. Hess, W. R. & Magnus, W. O. C. : Leck-und Kau-Automatismen bei electrischer Reizung am Swischenhirn, *Helv. Physiol. Acta*, 1943, I, pp. 535–547.

66. Hesse, E. & Reichelt, H. : Zur Wertbestimmung der Analgetika und ihre Kombination III. Mitteilung, *Arch. f. exp. Pathol. u. Pharmakol.*, 1933, *169*, p. 453.

67. Hewer, A. J. H. & Keele, C. A. : A Method of Testing Analgesics in Man, *Lancet*, 1948, *255*, pp. 683–688.

68. Hewer, A. J. H. & Keele, C. A., Keele, K. D. & Nathan, P. W. : Clinical Methods of Assessing Analgesics, *Lancet*, 1949 *256*, pp. 431–435.

69. Hildebrandt, F. : Die Prüfung der Analgetika im Tierexperiment mittels einer neuen Methode, *Arch. f. exp. Pathol. u. Pharmakol.*, 1934, *174*, pp. 403–415.

70. Hollander, H. L. & Gross, E. G. : The New Analgesics, *Fed. Proc.*, 1946, 7, pp. 227–228.

71. Houde, R. W. & Wikler, A. : Delineation of the Skin-twitch Response in Dogs and the Effects thereon of Morphine, Thiopental and Mephenesin, *J. Pharmacol.*, 1951, *103*, pp. 236–242.

72. Houde, R. W. & Wikler, A., Irwin, S. : Comparative Actions of Analgesic Hypnotic and Paralytic Agents on Hindlimb Reflexes in Chronic Spinal Dogs, *J. Pharmacol*, 1951, *103*, pp. 243–248.

73. Hougs-Olsen, W. : Comparative Investigations into the Analgesic Effects of Morphine and d,1-6-dimethylamino-4, 4-diphenyl-3-heptanone hydrochoride (Methadone, Butalgine), *Acta pharmacol & toxicol*, 1949, *5*, pp. 33–44.

74. Ipsen, J. Jr. : Biometric Analysis of Graded Response with Incomplete Measurements in Assays of Analgetic Drugs, *Acta Pharmacol & toxicol*, 1949, *5*, pp. 321–340.

75. Irwin, S. R., Houde, R. W., Bennett, D. R., Hendershot, L. C., Seevers, M. H. : The Effects of Morphine, Methadone and Meperidine on some Reflexes Responses of Spinal Animals to Nociceptive Stimulation, *J. Pharmacol.*, 1951, *101*, pp. 132–143.

76. Isbell, H., Eisenman, A. J. & Wikler, A. & Frank, K. : The Effects of Single Doses of 6-dimethylamino-4-4-diphenyl-3-heptanone (Amidone, Methadone or 10820) on Human Subjects, *J. Pharmacol.*, 1948, *92*, pp. 83–89.

77. Isbell, H., Wikler, A., Eisenman, A. J., Daingerfield, M. & Frank, K. : Liability of Addiction to 6-dimethylamino-4-4 diphenyl-3-heptanone (Methadone, Amidone or 10820) in Man, *Arch. Int. Med.*, 1948, *82*, pp. 362–392.

78. Isbell, H. & Vogel, V. H. : The Addiction Liability of Methadone (Amidone, Dolophine, 10820) and its Use in the Treatment of the Morphine Abstinence Syndrome, *Am. J. Psychiatr.*, 1949, *105*, pp. 909–914.

79. Isbell, H. & Fraser, H. F. : Addiction to Analgesics and Barbiturates, *Pharmacol. Rev.*, 1950, *2*, pp. 355–397.

80. Jacob, J., Ambrus, J. & Ambrus, C. : Effets analgésiques des administrations répétées de bichlorhydrate d'histamine chez le rat, *Ann. Institut Pasteur*, Paris, 1951, *81*, pp. 281–292.

81. Jacob, J. & Grassi-Gialdroni, G. : Influence des stimuli thermo-algésiques sur la réactivité à la douleur de la souris, *Experientia*, 1953, *9*, pp. 264–269.

82. Jacob, J. : L'Action analgésique de la morphine — Sa détermination expérimentale et son analyse pharmacologique, *Actualités Pharm.*, 1954, 7me série, pp. 127–171.

83. Jacob, J. : Pharmacodynamie de la douleur — La morphine, in *Douleur*, Alajouanine, Paris, 1956 (in the press).

84. Jackson, H. : The Evaluation of Analgesic Potency of Drugs, using Thermal Stimulation in the Rat, *Brit. J. Pharmacol*, 1952, 7, pp. 196-203.

85. Jackson, H. : The Effect of Analgesic Drugs on the Sensation of Thermal Pain in Man, *Brit. J. Pharmacol.*, 1952, 7, pp. 204–214.

86. de Jongh, D. K. & Knoppers, A. Th. : Action of Quinine Hydrochloride on Pain Thresholds in Animals, *Arch. Int. Pharmacol.*, 1952, *105*, 4, pp. 449–461 and 466–477.

87. Jores, A. & Frees, J. : Die Tagesschwinkungen der Schmerzempfindung, *Deutsche Med. Wchnschr.*, 1937, *63*, p. 962.

88. Keats, A. S. & Beecher, H. K. : Analgesic Potency and Side-action Liability in Man of Heptazone, in 1161-2,6-methyl Dihydromorphine, Metopon, Levo-isomethadone and Pentobarbital Sodium, as a Further Effort to refine Methods of Evaluation of Analgesic Drugs, *J. Pharmacol.*, 1952, *105*, 2, pp. 109–129.

89. KELLER, A. D. : Autonomic Discharges elicited by Physiologic Stimuli in Mild-brain Preparations, *Amer. J. Physiol.*, 1932, *100*, pp. 576–586.

90. KIESIG, H. J. & ORZECHOWSKI, G. : Untersuchungen über die Wirkungsweise der Sympathicomimetica, *Arch. f. exp. Pathol. u. Pharmakol.*, 1941, *197*, p. 391.

91. KOLL, W. & REFFERT, H. : Eine neue Methode zur Messung Analgetischer Wirkungen im Tierversuch, *Arch. f. exp. Pathol. u. Pharmakol.*, 1938, *190*, pp. 687–711.

92. KOLL, W. & FLEISHMANN, G. : Messungen der analgetischen Wirksamkeit einiger Antipyretica am Hund, *Arch. f. exp. Pathol. u. Pharmakol.*, 1941, *198*, pp. 390–406.

93. KOLL, W. : Messung analgetischer Wirkungen durch elektrische Zahnpulparcizung am Hund, *Arch. f. exp. Pathol. u. Pharmakol.*, 1952, *216*, pp. 83–89.

94. KRAUSHAAR, A. : Zur Wirkunganalyse Analgetischer Substanzen, *Arzneim. -Forsch.*, 1953, *3*, pp. 247–251.

95. KRUGER, H., EDDY, N. & SUMWALT, M. : The Pharmacology of the Opium Alkaloids, *Public Health Report*, 1940, suppl. *165*, parts 1 and 2.

96. KUHN, R. A. & BROMILEY, R. B. : Human Pain Thresholds determined by the Radiant Heat Technique and the Effect upon them of Acetylsalicylic Acid, Morphine Sulphate and Sodium Phenobarbital, *J. Pharmacol.*, 1951, *101*, pp. 47–55.

97. LEIMDORFER, A. : An Electroencephalographic Analysis of the Action of Amidone, Morphine and Strychnine, on the Central Nervous System, *Arch. int. Pharmacod.*, 1948, *76*, pp. 153–162.

98. LEIMBACH, D. & EDDY, N. : Synthetic Analgesics. — IV. Methadols, Isomethadol and their Acyl Derivatives, *J. Pharmacol.*, 1954, *110*, pp. 135–148.

99. LESPAGNOL, A., MERCIER, F., BERTRAND, Y. & MERCIER, J. : Recherches dans la série des Analgésiques, *Ann. pharm. franç.*, 1950, *8*, pp. 241–261.

100. LEWIS, J. : The Development of Tolerance in Rats to some New Synthetic Analgesics, *J. Pharmacol.*, 1949, *96*, pp. 31–37.

101. LEWIS, Th., PICKERING, G. W. & ROTSCHIL, P. : Observations upon Muscular Pain in Intermittent Claudication, *Heart*, 1931, *15*, p. 359.

102. LEWIS, Th. : Pain in Muscular Ischemia : its Relation to Anginal Pain, *Arch. Int. Med.*, 1932, *49*, p. 713.

103. LEWIS, Th. : Pain, McMillan, New York, 1942.

104. L'HERITIER, Ph. : Les méthodes statistiques dans l'expérimentation biologique, *C.N.R.S.*, Paris, 1949.

105. LINDER, A. : Sur la manière d'organiser les expériences afin d'obtenir un rendement maximum, *Arch. Sc. Phys. Nat.*, 1946, *28*, pp. 181–191.

106. LIBMAN E. : Observations on Sensitiveness to Pain, *Tr. A. Am. Physicians*, 1926, *41*, p. 305.

107. LITCHFIELD, J. T. Jr. & WILCOXON, F. : A Simplified Method of Evaluating Dose-effect Experiments, *J. Pharmacol.*, 1948, *96*, pp. 99–113.

108. LITCHFIELD, J. T. Jr. & WILCOXON, F. : The Reliability of Graphic Estimation of Relative Potency from Dose per cent Effect Curves, *J. Pharmacol.*, 1953, *108*, pp. 18–25.

109. LUCKARDT, A. B. & JOHNSON, C. A. : Studies on the Knee Jerk. IV. The Effect of Moderate Doses of Morphine Sulphate on the Knee Jerk of the Cat, *Amer. J. Physiol.*, 1928, *83*, p. 653.

110. MAGUN, R. : quoted by Soehring, 1947, 1948.

111. MAZOUE, H. : Modifications des lois de sommation sous l'influence d'application de morphine sur la moëlle, *C.R. Soc. Biol.*, 1927, *97*, p. 465.

112. MCGOWAN, J. M., BUTSCH, W. L. & WALTERS, W. : Pressure in the Common Bile Duct of Man, its Relation to Pain following Cholecystectomy, *J.A.M.A.*, 1936, *106*, p. 2227.

113. MILLER, L. C. & TAINTER, M. L. : Estimation of ED_{50} and its Error by means of logarithm-probit Graph-paper, *Proc. Soc. Exper. Biol. Med.*, 1944, *261*, p. 57.

114. MILLER, L. C. : A Critique of Analgesic Testing Methods, *Ann. N. Y. Acad. Sc.*, 1948, *51*, 1, pp. 34–50.

114*b*. MONNIER, M. : *XXth Internat. Physiol. Congr., Brussels*, July-Aug. 1956.

115. MUMMERY, J. W. : Nerve Supply of the Dentine, *Proc. Roy. Soc. Med.*, 1924, *17*, p. 35.

116. MUNCH, J. : Untersuchungen über die Innervierung der menschlichen Zahn-pulpa und des Dentins, *Wiertel. Zahnh.*, 1927, *43*, p. 503.

117. OHLSSON, L. : Algesimetric Studies by the Hot-plate Method in Mice, *Acta Pharmacol. toxicol.*, 1953, *9*, pp. 322–331.

118. NIMS, L. F., MARSHALL, C. & NIELSEN, A. : Effects of Local Freezing on the Electrical Activity of the Cerebral Cortex, *Yale J. Biol. & Med.*, 1941, *13*, 477–484.

119. PETERSEN, P. : Studies on a new Spasmolytic Compound 1,1-diphenyl-3-dimethylaminobutene (A 29), related to Methadone and on the Combined Use of this Compound and a Potent Analgesic, Keto-bemidone, *Acta pharmacol. toxicol.*, 1951, *7*, pp. 51–64.

120. PFEIFFER, C. C., SONNENSCHEIN, R. R., GLASSMAN, L., JENNEY, E. H., BOGOLUB, S. : Experimental Methods for Studying Analgesia, *Ann. N.Y. Acad. Sc.*, 1948, 51, *1*, pp. 21–33.

121. RADOUCO-THOMAS, C., FROMMEL, Ed., GOLD, Ph., GREDER, G., MELKONIAN, D., RADOUCO-THOMAS, S., STRASSBERGER, L., VALLETTE, F. & DUCOMMUN, M. : La physiologie de l'épilepsie électrique expérimentale, *Arch. int. pharmacodyn.*, 1952, 92-2, 129–162.

122. RADOUCO-THOMAS, C. : *Epilepsie et médication antiépileptique. Physio-Pathologie, pharmacologie et thérapeutique*, Georges. Geneva, 1956.

123. RADOUCO-THOMAS, C. & RADOUCO-THOMAS, S. : Morphine, morphinomimétiques et morphinoblocants, *Arch. Sc.*, Geneva, 1956, *9*, 1, pp. 117–123.

124. RADOUCO-THOMAS, C., RADOUCO-THOMAS, S. & NOSAL, Gl. : Contribution to the Study of Synthetic Analgesic Narcotics, *Bulletin on Narcotics*, 1956, VIII. 2, pp. 18–25.

124*b*. RADOUCO-THOMAS, C. : Morphine et morphinomimétiques en anesthésie générale, *Congrès d'Anesthésiologie*, Zurich, Aug. 1956.

125. RADOUCO-THOMAS, C., RADOUCO-THOMAS, S. & Ed. FROMMEL : Etude comparative de l'activité analgésique de la phénylbutazone, de l'amidopyrine et de la morphine, *Réunion Annuelle des Physiologues et Pharmacologues*, Fribourg, mai 1956, *Helv. Physiol. Acta* (sous presse).

126. RADOUCO-THOMAS, C., RADOUCO-THOMAS, S. & NOSAL, Gl. : Psycho-neuroses expérimentales engendrées par la morphine et les morphinomimétiques. Effet des morphinoblocants, de la médication psychotonique et psychoplègique (barbituriques, chlorpromazine, réserpine et bénactyzine). *Congrès Intern. de Physiologie*, Bruxelles, août 1956.

127. RADOUCO-THOMAS, S. : Contribution à la pharmacodynamie et au Mode d'Action des anticonvulsivants. *La Diphénylhydantoine*, thèse biol. et chim. med., Geneva, 1952.

128. RADOUCO-THOMAS, S. : Thèse Sci., Paris 1957 (en préparation).

128*b* RANDALL, L. & LEHMANN, G. : Pharmacological Studies on Analgesic Piperidine Derivatives, *J. Pharmacol.*, 1948, *93*, p. 3114.

129. RICKARDS, J. C., BOXER, G. E. & SMITH, C. C. : Studies on the Distribution and Metabolism of Methadone in Normal and Tolerant Rats by a new calorimetric method, *J. Pharmacol.*, 1950, *98*, p. 380.

130. RISING, L. W. & LYNN, E. V. : A Toxicological Investigation of Morphine and Codeine, *J. Am. Pharm. Ass.*, 1932, *21*, p. 334.

131. RUBSAMEN, W. : Experimentelle Untersuchungen über die Gewöhnung an Morphine, *Arch. f. exp. Pathol. u. Pharmakol.*, 1908, *59*,. p. 227.

132. RUCKSTHUL, K. : *Beitrag zur Pharmacodynamischen Prüfung der Analgetica*. Dissertation, Bern, 1939.

133. SCHAUMANN, O. : Die neuen Synthetischen Analgetika, *Arch. f. exp. Pathol. u. Pharmakol.*, 1952, *216*, pp. 48–77.

134. SCHERRER, J., BARBIZET, J. & OUDOT-LE HENAFF, M. : Contribution à l'étude de l'organisation temporo-spatiale de la sensibilité somesthétique normale, *Année Psychologique*, 1954, *1*, pp. 53–81.

135. SCHERRER, J., MARTY, R., PERTUISET, F. & CONTAMIN, F. : Effets sur l'électro-corticogramme du lapin de stimulations sensitives, thermiques et nociceptives, *C.R. Séances de l'Acad. Sc.*, Paris, 1955, *241*, pp. 247–249.

136. SCHERRER, J. : A propos de quelques aspects physiologiques de la douleur, in *Douleur*, Alajouanine, Paris, 1956 (in the press).

137. SCHOEN, R. : Klinische Stellung zu den Analgetica, *Arch. f. Exp. Pathol. u. Pharmakol.*, 1953.

138. SCHROEDER, S. : Der Induktionsstrom als Diagnostikum in der Zahnärztlichen Praxis, *Deutsche Zahnklinik*, 1907, *11*, p. 83.

139. SCHAUMACHER, G. A., GOODELL, H., HARDY, J. D. & WOLFF, H. G. : Uniformity of the Pain Threshold in Man, *Science*, 1940, *92*, pp. 110–112.

140. SEEVERS, M. H. & PFEIFFER, C. C. : J. Pharmacol., 1936, *56*, p. 166.

141. SEEVERS, M. H., BENNET, J. H., POHLE, H.W. & REINARDY, E. W. : J. Pharmacol., 1937, *59*, p. 291.

142. SINNIGER, A. : *Un antithermique et analgésique nouveau : le 1610. — Etude expérimentale*, medical thesis, Geneva, 1953.

143. SMITH, C. & LEHMAN, E. : Analgesics — I, Effects of Analgesic Combinations on Reaction Time in Rat, *J. Pharmacol.*, 1953, *108*, pp. 336–339.

144. SOEHRING, K. & BECHER, F. : Über die Messung Analgetischer Wirkungen im Tierversuch, *Arch. int. Pharmacodyn.*, 1949, *79*, pp. 45–69.

145. SONNENSCHEIN, R. R. & IVY, A. C. : Failure of Oral Antipyretic Drugs to alter Normal Human Pain Threshold, *J. Pharmacol.*, 1949, *97*, 308–313.

146. STARKENSTEIN, E. : Zur Methodik der Prüfung Schmerzstillender Mittel im Tierversuch, *Arch. f. exp. Pathol. u. Pharmakol.*, 1932, *165*, pp. 325–338.

147. STEINHAUSEN, W. : Uber Stromdichtebestimmung und die Beziehung der Stromdichte zum Erregungsvorgang, *Pflüngers Arch.*, 1921, *193*, pp. 171–200.

148. STELLMACH, R.: *Inaugural-Diss.*, Pharmakol. Inst. Univ., Hamburg, 1948.

149. STELLMACH, R., SCHRODER, E. & SOEHRING, K. : Uber die Messung Analgetischer Wirkungen im Tierversuch. III. : Quantitative Auswertung von Lokalanalgetica am Hundeschädel, *Arch. int. pharmacodyn.*, 1952, 89, pp. 380–398.

150. STRAUB, W. & TRIENDLE, E. : Über die Messung analgetischersedativer Wirkungen am Tier und die Übertragbarkeit der Messungsresultats auf den Menschen, *Arch. f. exp. Pathol. u. Pharmakol.*, 1940, *195*, pp. 481–490.

151. SUNG, C. Y. & WAY, E. L. : The Effect of the Liver on the Metabolism of d-l-Methadone in vitro and in vivo., *J. Pharmacol.*, 1950, *98*, p. 72.

152. Symposium on Pain — *Association for Research in Nervous and Mental Diseases* (Volume XXIII) Pain, Williams & Wilkins Co., Baltimore, 1943.

153. Symposium : Newer Synthetic Analgesics — *Ann. N.Y. Acad. Sc.*, 1948, *51*, 2, pp. 1–174.

154. THORP, R. H. : The Assessment of Analgesic Activity in New Synthetic Drugs, *Brit. J. Pharmacol.*, 1946, *1*, pp. 113–126.

155. THORP, R. H. : The Pharmacology of the Optical Isomers of Amidone (2-dimethylamoni-4-4-diphenylheptan-5-one), *Brit. J. Pharmacol.*, 1949, *4*, pp. 98–104.

156. THORP, R. H., WALTON, E. & OFNER, O. : *Nature*, 1947, *159*, p. 679.

157. TOMAN, J. E. P. & DAVIS, J. P. : The Effects of Drugs upon the Electrical Activity of the Brain, *J. Pharmacol.*, 1949, *97*, 2, pp. 425–492.

158. TYE, A. & CHRISTENSEN, B. : The Estimation of some Analgetic Potencies, *J. Amer. Pharm. Assoc.*, 1952, *41*, pp. 75–77.

159. VESSEREAU, A. : *Méthodes statistiques en biologie et en agronomie*, Baillière, Paris 1948.

160. WERZ, R. M. : Über die Chronaxie der sensiblen Nervendigungen im Zahnbein, *Arch. f. exp. Pathol. u. Pharmakol.*, 1932, *167*, pp. 191–195.

160*b*. WHITE & SWEET : *Pain*, C. C. THOMAS, Springfield (Ill.), 1955.

161. WIKLER, A. : Sites and Mechanisms of Action of Morphine and Related Drugs in the Central Nervous System, *Pharmacol., Rev.*, 1950, Vol. 2, 3, pp. 435–506.

162. WIKLER, A. : *Opiate Addiction : Psychological and Neurophysiological Aspects in Relation to Clinical Problems*, Ch. Thomas, Springfield, Ill., 1953.

163. WIKLER, A., GOODELL, H. & WOLFF, H. G. : Studies on Pain : The effects of analgesic agents on sensations other than pain, *J. Pharmacol.* 1945, *83*, p. 4.

164. WILHEMI, VON G. : Über die Pharmakologisches Eigenschaften von Irgapyrineinen neuen Preparat aus der Pyrazolreihe, *Schweiz. Med. Wschrift.*, 1949, *79*, p. 577.

165. WINDER, C. V., PFEIFFER, C. C. & MAISON, G. L. : The Nociceptive Contraction of the Cutaneous Muscle of the Guinea-pig as elicited by Radiant Heat, with Observations on the Mode of Action of Morphine, *Arch. int. Pharmacod.*, 1946, 72, 3-4, pp. 329–359.

166. WINDER, C. V. : A Preliminary Test for Analgetic Action in Guinea Pigs, *Arch. int. Pharmacodyn.*, 1947, *74*, 2, pp. 176–192.

167. WINDER, C. V. : Quantitative Evaluation of Analgetic Action in Guinea Pigs — Morphine, Ethyl 1-methyl-4-phenylpiperidine-4-carboxylate (demoral and Acetylsalicylic Acid, *Arch. int. Pharmacodyn*, 1947, *74*, 3-4, pp. 219–232.

168. WINDER, C. V. : Distribution of the Resting Pain-reaction Thresholds in Guinea-pigs, with a statistical Concept of gradation of biological Effect, *Yale J. Biol. Med.*, 1947, *19*, pp. 289–310.

169*a*. WINTER, C. A. & FLATAKER, L. : The Relation between Temperature and the Effect of Morphine upon the Response to Thermal Stimuli in the Albino Rat and the Dog, *J. Pharmacol.*, 1953, *109*, 1, pp. 183–188.

169*b*. WINTER, C. A. & FLATAKER, L. : Studies on Heptazone (6-Morpholino-4-4-diphenyl-3-heptazone hydrochloride) in comparison with other Analgesic Drugs, *J. Pharmacol.*, 1950, *98*, pp. 305–317.

170. WIRTH, W. : Methadon der Analgesioprüfung am Tier, *Arch. f. exp. Pathol. u. Pharmakol.*, 1952, *216*, p. 77.

171. WOLFF H. G., HARDY, J. D. & GOODELL, H. : Studies on Pain. Measurements of the Effect of Morphine, Codeine and Other Opiates on the Pain Threshold and an Analysis of their Relation to the Pain Experience, *J. Clin. Invest.*, 1940, *19*, p. 659.

172. WOLFF, H. G., HARDY, J. D. & GOODELL, H. : Measurements of the Effect on the Pain Threshold of Acetylsalicylic Acid, Acetanilid, Acetophenetidine, Aminopyrine Ethyl Alcohol, Trichloroethylene, a Barbiturate, Quinine, Ergotamine tartrate and Caffeine : an analysis of their Relationship to the Pain Experience, *J. Clin. Invest.*, 1941, *20*, p. 63.

173. WOLFF, H. G., HARDY, J. D. & GOODELL, H. : Studies on Pain : Measurements of the Effect of Ethyl Alcohol on the Pain Threshold and on the " Alarm " Reaction., *J. Pharmacol.*, 1942, *75*, p. 38.

174. WOLFF, H. G. & HARDY, J. D. : On the Nature of Pain, *Physiol. Rev.*, 1947, *27*, pp. 167–199.

175. WOLFF, H. G. & WOLFF, S. : *Pain*, Ch. Thomas, Springfield, Ill., 1951.

176. WOOLFE, G. & MACDONALD, A. D. : The Evaluation of the Analgesic Action of Pethidine Hydrochloride (Demerol), *J. Pharmacol.*, 1944, *80*, pp. 300–307.

177. WRIGHT, C. I., BARBOUR, F. A. : The Respiratory Effects of Morphine, Codeine, and Related Substances. III. The Effect of Morphine, Dihydromorphine, Dihydromorphinone (Dilaudid) and Dihydrocodeinone (Dicodid) on the respiratory Activity of the Rabbit, *J. Pharmacol.*, 1935, *53*, pp. 34–45.

178. YIM, G. K. W., KEASLING, H. H., GROSS, E. G. & MITCHELL, C. W. : Simultaneous Respiratory Minute Volume and Tooth Pulp Threshold Changes, following Levorphan, Morphine and Levorphan-Levallorphan mixtures in Rabbits, *J. Pharmacol.*, 1955, *115*, 1, pp. 96–105.

179. YONKMAN, F. F., NOTH, P. H. & HECHT, H. : Some Pharmacological Features of Demerol, *Proc. Central Soc. Clin. Research*, 1942, *15*, p. 89.

180. ZEROSI, C. : Sull' Innervazione del Dente et del Paradenzion, *Rass. trimest. Odont.*, 1946, *2*, p. 67.

Quantitative Determination of Morphine in Opium by Paper Chromatography and Spectrophotometry

By Haruyo Asahina and Masako Ono
National Hygienic Laboratory, Tokyo, Japan

Introduction

Paper chromatography was used with good results by the authors for the determination of morphine in opium tincture.*

By this method, we now estimate the morphine content in opium. As only a small quantity of opium is employed, it is possible to determine the morphine content of opium from one poppy capsule—i.e., between 0.03 and 0.5 g of opium per poppy head, and to compare the morphine content in opium from successive lancings of the same poppy capsules. We have confirmed the decline in morphine content with successive lancings.

Procedure

Extraction

The extraction of morphine with hydrochloric acid was found to be satisfactory. The other opium alkaloids and the coloured substances were easily separated from morphine in the paper-chromatographic step.

About 0.1 g of air-dried opium was accurately weighed and 1 cc. of N/10 hydrochloric acid (precisely measured) was added, and the mixture was rubbed with a glass rod to a homogeneous paste. The resulting suspension was centrifuged at 2,000 r.p.m. for thirty minutes and 0.003 cc. of the supernatant solution was measured by a micro-pipette with accuracy.

Chromatography

Chromatography of an opium tincture made to contain 1% morphine was carried out in the same container, in order to obtain a calibration curve. In this case, 0.001, 0.003, 0.006 cc of opium tincture (10 γ, 30 γ, 60 γ of morphine) was applied to the paper.

Solvent : n-butanol 50, 28% ammonia 9, distilled water 15 parts by volume. The top layer of the mixture, 40 cc. for each container, was used.

Paper : Toyo No. 51, 2.5 cm × 40 cm.

Container : inner diameter 9.5 cm., height 45 cm.

Starting line : 7 cm from the lower edge of paper.

At the start of the chromatography, about 0.5 cm. of paper dipped in the solvent.

Height of development : approximately 25 cm. above the starting line.

Measurement of Absorbance by Beckman Spectrophotometer

The maximum absorbances of the morphine spots on the paper of both the opium tincture and the opium extract were determined at 286 mμ.

Recently we have devised a simple apparatus for passing the filter-paper which is more convenient than the one previously used.[1] In place of the sample chamber, an accessory of our making (fig. 1) is attached tightly to the Beckman spectrophotometer DU by using the part of the accessory for fluorescence.

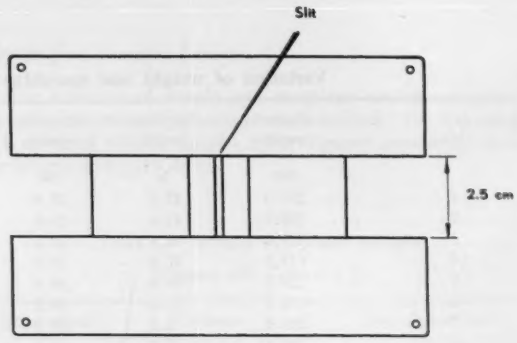


FIGURE 1. *Front view* (scale ½)

Through A of our accessory, the filter paper is passed horizontally (fig. 2). The strips of filter paper on which the chromatograms are developed are 2.5 cm wide. The height of A is 2.5 cm—that is, the width of the strip of filter-paper. The thickness of the slot A is just a little more than that of the filter-paper.

After development with solvent in the chromatography, the strip of filter-paper is fastened to the end of a celluloid plate (length about 25 cm, width exactly 2.5 cm) with cello-tape (fig. 3), and this plate is used to pull the strip of filter-paper through the apparatus.

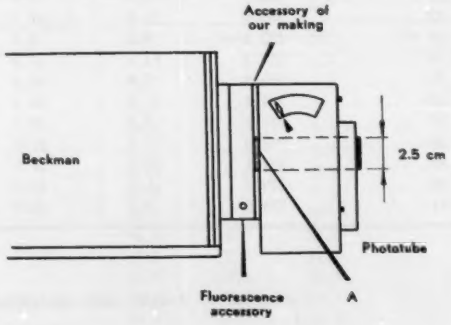


FIGURE 2. *Side view*

* See Document ST/SOA/SER.K/40, 25 August 1955.

[1] See United Nations document ST/SOA/Ser. K/40.

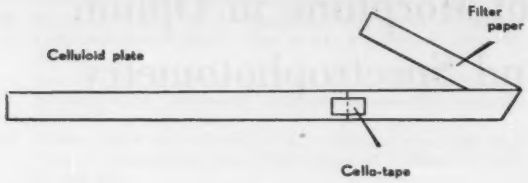


FIGURE 3

Pulling the celluloid plate slowly by hand, the place of maximum morphine absorbance on the filter-paper is searched for, while the absorbance of the other part of the filter-paper (where there is no spot) is set at zero.

Calculation

From the calibration curve of the opium tincture and the absorbance of the morphine spot of the opium extract thus obtained, the quantity of morphine in 0.003 cc. of the opium extract was determined, and from it the content of morphine in the opium was calculated.

TABLE A

Variation of weight and morphine content of opium from individual capsules

No.	*Opium weight*	*Morphine content*	*Morphine quantity*	*No.*	*Opium weight*	*Morphine content*	*Morphine quantity*
	mg	%	mg		mg	%	mg
1	262.3	13.7	35.9	42	296.3	10.5	31.1
2	105.0	12.4	13.0	43	272.1	10.5	28.6
3	219.0	12.4	27.2	44	227.3	15.3	34.8
4	173.9	11.3	19.7	45	154.2	4.5	6.9
5	229.2	15.0	34.4	46	160.9	11.3	18.2
6	171.8	11.2	19.2	47	277.7	11.9	33.0
7	226.9	5.2	11.8	48	81.9	11.4	9.3
8	94.4	7.1	6.7	49	298.1	18.1	54.0
9	115.1	12.1	13.9	50	88.3	11.5	10.2
10	184.0	12.5	23.0	51	231.7	12.4	28.7
11	88.2	10.2	9.0	52	124.7	11.3	14.1
12	281.5	10.9	30.7	53	201.0	13.2	26.5
13	130.0	10.2	13.3	54	163.7	12.3	20.1
14	223.9	13.8	30.9	55	92.4	9.3	8.6
15	218.6	12.5	27.3	56	184.3	13.4	24.7
16	54.4	11.3	6.1	57	285.1	13.6	38.8
17	275.7	12.2	33.6	58	219.5	12.9	28.3
18	121.2	15.3	18.5	59	186.7	9.7	18.1
19	186.4	9.7	18.1	60	80.0	17.7	14.2
20	420.8	15.5	65.2	61	59.8	12.0	7.2
21	220.0	9.8	21.6	62	34.8	13.9	4.8
22	136.4	12.9	17.6	63	173.5	9.4	16.3
23	270.1	12.6	34.0	64	95.5	14.3	13.7
24	267.3	12.8	34.2	65	138.1	9.5	13.1
25	469.5	6.4	30.0	66	134.0	6.6	8.8
26	243.4	12.4	30.2	67	172.0	8.9	15.3
27	202.7	13.7	27.8	68	77.7	9.1	7.1
28	192.0	11.0	21.1	69	52.3	13.3	7.0
29	240.9	14.5	34.9	70	154.1	9.2	14.2
30	139.4	8.2	11.4	71	162.6	9.3	15.1
31	106.0	7.0	7.4	72	154.5	10.3	15.9
32	363.4	11.3	41.1	73	158.0	7.8	12.3
33	122.6	5.1	6.3	74	31.7	16.3	5.2
34	215.3	12.7	27.3	75	36.6	12.1	4.4
35	126.9	9.9	12.6	76	223.6	12.3	27.5
36	244.4	11.7	28.6	77	283.7	12.6	35.7
37	247.2	7.9	19.5	78	79.9	14.5	11.6
38	291.9	7.4	21.6	79	126.2	11.8	14.9
39	271.1	13.7	37.1	80	163.4	8.7	14.2
40	279.4	15.7	43.9	81	85.9	7.7	6.6
41	278.6	9.3	25.9	82	305.2	10.9	33.3

TABLE B

Variation of weight and morphine content of opium from individual capsules

Poppy plant	Japanese origin, Ikkanshu variety
Crop season	1954–55
Cultivator	Kasukabe Botanical Garden
Method of morphine determination	paper chromatography and spectrophotometry

TABLE B (*continued*) — **Number of capsules in different classes**

Opium weight		*Morphine content*		*Morphine quantity*	
Below 100 mg	16	Below 5%	1	Below 10 mg	16
100-200	30	5-10	24	10-20	28
200-300	32	10-15	49	20-30	17
Above 300	4	Above 15	8	30-40	17
				40-50	2
TOTAL ...	82	TOTAL ...	82	Above 50	2
				TOTAL ...	82

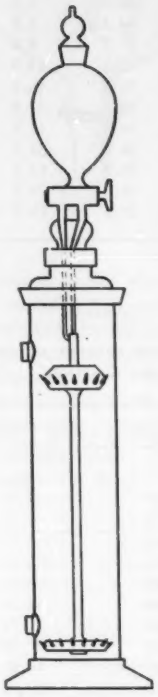


FIGURE 4

Chromatographic chamber

Opium from Successive Lancings

The variation of weight and morphine content of opium samples from successive lancings was examined. The following 15 air-dried opium samples from *Papaver somniferum* were produced during 1954–55.

TABLE 1 — **Origin of opium sample**

(Compare with Tables 2 to 7)

Opium sample	*Cultivator*	*Seed obtained from*
1	Kasukabe Botanical Garden	Rome, Italy
2	"	Adelaide, Australia
3	"	Amsterdam, Netherlands
4	"	Torino, Italy
5	"	Braunschweig, Germany
6	"	Amsterdam, Netherlands
7	"	London, England
8	"	Adelaide, Australia
9	"	Amsterdam, Netherlands
10	"	Torino, Italy
11	Drug company in Kyoto	Pakistan
12	"	Japan, ordinary
13	"	Pakistan
14	Wakayama Botanical Garden	Ikkanshu, Japan
15	"	" "

TABLE 2

Weight of opium from successive lancings

(In grammes)

Opium sample	*Lancing*				
	1st	*2nd*	*3rd*	*4th*	*5th*
1	0.7642	0.2958			
2	0.6773	0.0668			
3	0.7105	0.3996	0.0742		
4	1.0974	0.6213	0.0608		
5	0.6590	0.5194	0.0744		
6	0.3746	0.1989	0.0351		
7	0.1488	0.2484	0.0577		
8	0.0314	0.0098	0.0202		
9	0.0400	0.0253	0.0253		
10	0.0721	0.0214	0.0176		
11	3.3026	5.4959	2.7627	3.1152	0.0969
12	0.1782	0.1179	0.1614		
13	0.2789	0.3300	0.1319		
14	22.7462	14.6107	6.7535	5.2203	
15	16.6977	15.6183	14.9554	12.3396	

TABLE 3

Percentage of weight from each lancing to total weight

Opium sample	*Lancing*				
	1st	*2nd*	*3rd*	*4th*	*5th*
1	72.1	27.9			
2	91.0	9.0			
3	60.0	33.7	6.3		
4	61.7	34.9	3.4		
5	52.6	41.5	5.9		
6	61.5	32.7	5.8		
7	32.7	54.6	12.7		
8	51.1	16.0	32.9		
9	44.2	27.9	27.9		
10	64.9	19.3	15.8		
11	22.3	37.2	18.7	21.1	0.7
12	38.9	25.8	35.3		
13	37.7	44.5	17.8		
14	46.1	29.6	13.7	10.6	
15	28.0	26.2	25.1	20.7	

TABLE 4

Morphine content of opium from successive lancings

(Percentage)

Opium sample	Lancing					Method used
	1st	2nd	3rd	4th	5th	
1	9.6	7.5				A
2	12.8	14.2				A
3	21.5	13.4	10.0			A
4	15.7	14.4	11.3			A
5	20.0	16.3	5.7			A
6	14.5	15.0	8.6			A
7	17.3	15.8	11.7			A
8	15.4	9.4	8.5			A
9	18.6	18.5	9.5			A
10	15.4	17.2	12.5			A
11	17.01	9.92	7.33	6.04	7.3	1st–4th, B; 5th, A
12	9.0	8.9	10.3			A
13	18.3	10.6	9.8			A
14	24.84	17.39	13.78	15.30		B
15	21.42	16.05	11.58	10.48		B

Reference

Machiguchi: *J. Pharm. Soc. Japan, 529,* 193 (1926)

1	22	20	17	14	
2	20	16	12	8	5

Method used for morphine in opium

A: Paper chromatography and spectrophotometry.

B: Slightly modified Japanese Pharmacopoeia VI using 1 g. of opium.

TABLE 5

Calculated morphine quantity

(In milligrammes)

Opium sample	Lancing				
	1st	2nd	3rd	4th	5th
1	73.36	22.19			
2	86.69	9.49			
3	152.76	53.55	7.42		
4	172.29	89.47	6.87		
5	131.80	84.66	4.24		
6	54.32	29.84	3.02		
7	25.74	39.25	6.75		
8	4.84	0.92	1.72		
9	7.44	4.68	2.40		
10	11.10	3.68	2.20		
11	561.77	545.19	202.51	188.16	7.07
12	16.04	10.49	16.62		
13	51.04	34.98	12.93		
14	5,650.2	2,540.8	930.6	798.7	
15	3,576.6	2,506.7	1,731.8	1,293.2	

TABLE 6

Percentage of morphine quantity from each lancing to total morphine

Opium sample	Lancing				
	1st	2nd	3rd	4th	5th
1	76.8	23.2			
2	90.1	9.9			
3	71.5	25.0	3.5		
4	64.1	33.3	2.6		
5	59.7	38.4	1.9		
6	62.3	34.2	3.5		
7	35.9	54.7	9.4		
8	64.7	12.3	23.0		
9	51.3	32.2	16.5		
10	65.4	21.7	12.9		
11	37.3	36.2	13.5	12.5	0.5
12	37.2	24.3	38.5		
13	51.6	35.3	13.1		
14	57.0	25.6	9.4	8.0	
15	39.3	27.5	19.0	14.2	

TABLE 7

Calculated morphine content of opium from all the lancings

Opium No.	Total quantity	Total morphine	Morphine content
	g	mg	%
1	1.0600	95.55	9.0
2	0.7441	96.18	12.9
3	1.1843	213.73	18.0
4	1.7795	268.63	15.1
5	1.2528	220.70	17.6
6	0.6086	87.18	14.3
7	0.4549	71.74	15.8
8	0.0614	7.48	12.2
9	0.0906	14.52	16.0
10	0.1111	16.98	15.3
11	14.7733	1,504.70	10.2
12	0.4575	43.15	9.4
13	0.7408	98.95	13.3
14	49.3307	9,920.3	20.1
15	59.6110	9,108.3	15.3
14+15	108.9417	19,028.6	17.5

Two Collections from each Lancing

The above samples 14 and 15 represent collections from the same capsules of the Ikkanshu variety, No. 14 the *Kiritori* collection and No. 15 the *Asabuki* collection.

In the *Kiritori* collection the unripe capsule is incised, and immediately afterward the latex which flows out is taken from the capsule. Its meaning is " cutting " (kiri) and " taking from " (tori).

The *asabuki* collection is made from the same lancing, and is the juice collected early the following morning. Its meaning is " flowing out " (buki) " at dawn " (asa).

The cultivators commonly mix the opiums obtained in these two collections, but they were kept separate in these trials.

Methods of Successive Incision

We have also made a comparison of opiums obtained by two different methods of successive incision, *setsuzoku-kiri* and *hantai-kiri*. The cultivator commonly incises every second day until no more latex flows from the capsule. This usually requires four lancings. Vertical incisions are made by means of a knife having three edges.

The ribs of the poppy capsules are popularly called " mountains ". In both methods, two adjacent ribs or " mountains " are incised at the first lancing. *Setsuzoku-kiri* (adjacent cutting), means that at the second lancing three ribs adjacent to the first two are incised, and so on around the capsule at the third and fourth lancings. *Hantai-kiri* (opposite cutting)

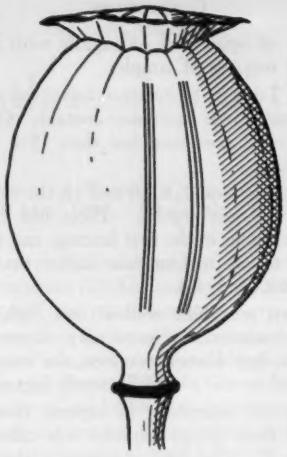


FIGURE 5. *Poppy head : first lancing*

means that at the second lancing three ribs adjacent to each other but opposite to the two already cut are incised. The third lancing is then made by incising three " mountains " between those cut in the first and second lancings, and the fourth lancing is then opposite to the third. *Setsuzoku-kiri* is usually used, as it is less laborious.

In these trials, poppy plants of the Ikkanshu variety were cultivated in two separate areas. The soil of area B was more fertile than that of area A.

No.	*Area*	*Method of incision*	*Number of capsules incised*	*Opium weight*
				g
1	A (poor soil)	Setsuzoku-kiri	135	15.2
2	A „ „	Hantai-kiri	129	13.7
3	B (rich soil)	Setsuzoku-kiri	132	15.6
4	B „ „	Hantai-kiri	110	11.2

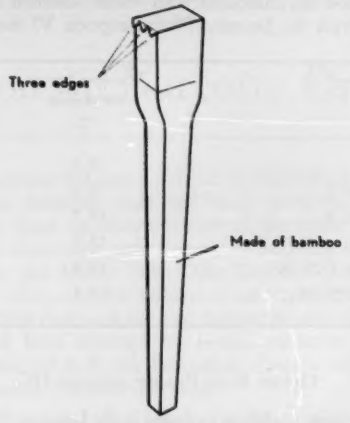


FIGURE 6. *Knife for incision*

No.	*Opium weight per capsule (Grammes)*	*Moisture content (Per cent)*	*Morphine percentage as determined (Per cent)*	*Morphine quantity per capsule (Grammes)*
1	0.113	10.53	11.18	0.0126
2	0.106	9.96	12.20	0.0130
3	0.118	10.84	13.36	0.0158
4	0.102	10.91	13.88	0.0141

The opium produced in the more fertile area had a higher morphine percentage. Differences between the results by the two methods of successive incision in these two trials are not considered sufficiently great to establish that they have any significance.

This year we shall study this problem in greater detail.

COMPARISON OF ASSAY RESULTS

In the following table, results of morphine determinations assayed on various samples of opium of different origin by

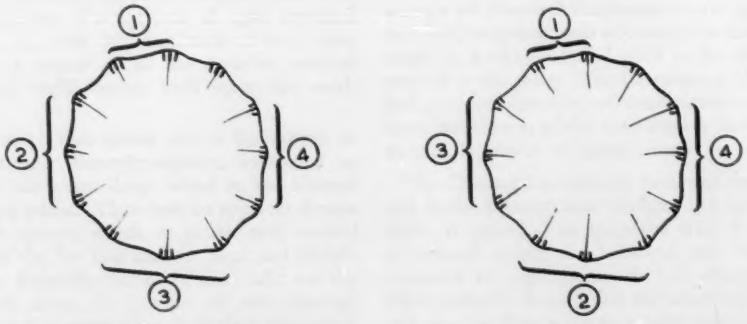


FIGURE 7. *Cross-section of poppy head*
Order of lancings

our method are compared with results obtained on the same samples with the Japanese Pharmacopoeia VI method.

Sample	*Morphine content our method* %	*J. P. VI* %
China	9.5	8.43
Illicit origin I	13.3	10.80
Illicit origin II	14.7	13.47
Turkey Export U.N. 15	13.5	13.61
India Excise U.N. 36	11.6	9.98
Iran Fars U.N. 47	12.8	11.22

Opium from *Papaver setigerum* DC

Papaver setigerum DC is included in the Japanese Opium Law. We obtained some seed by the courtesy of Dr. Charles G. Farmilo, Organic Chemistry and Narcotic Section, Food and Drug Laboratories, Department of National Health and Welfare, Ottawa, Canada, and cultivated this poppy experimentally at the Kasukabe Experiment Station of Medicinal Plants attached to the National Hygienic Laboratory, Saitama, Japan.

The opium from *Papaver setigerum* gave the following results on analysis :

Morphine content by our method of paper chromatography and spectrophotometry for morphine in opium, 5.1%;

Porphyroxine-meconidine absorbance, Beckman B, 1 cm cell, 1% solution, 0.235.

The presence of codeine and other opium by-alkaloids was demonstrated by the paper chromatogram.

Conclusions

The quantity of opium fell off rapidly with later incisions in a substantial number of samples.

As shown in Table 4, 11 opium samples out of 15 showed a marked decline in the morphine content. Of these, only one sample, No. 1, contained less than 15% of morphine in the first lancing.

In opium samples Nos. 2, 6, 10 and 12, the change of morphine content was not regular. These had comparatively low morphine content in the first lancing, and the morphine content of the second incision was higher than that of the first incision (Nos. 2, 6, 10).

When the first morphine content was high, a large and steady decline occurred, as previously shown by Annett and Machiguchi; but when it was low, the second morphine content increased in comparatively small degree.

The quantity of morphine in opium from successive lancings of the same poppy capsules was calculated and is given in Table 5. The figures generally show a definite diminution of morphine quantity with repeated incisions, in most of these opium samples.

It was thus possible to demonstrate that the chief reason for the lower morphine content of Japanese opium as compared with that of Turkey, Yugoslavia, Greece, and Bulgaria, where it is customary to bleed each capsule only once, is the successive scarifications of the poppy capsules in Japan.

For example, the first morphine content of Japanese opium (Ikkanshu), Nos. 14, 15, was more than 20%. But usually in our country, the capsules are incised repeatedly, four times on different days, and all the opium so obtained is mixed together. The mixed opium sample, Nos. 14, 15, contained 17.5% of morphine, whereas the morphine content of the first opium from these capsules was 24.8% and 21.4% respectively.

OFFICIAL

Eleventh session of the Commission on Narcotic Drugs

The Commission on Narcotic Drugs met in Geneva from 23 April to 18 May 1956. At the first meeting it elected as officers :

Mr. J. H. WALKER (United Kingdom), Chairman,
Mr. H. J. ANSLINGER (USA), (in absentia), Vice-Chairman
Dr. Amin ISMAIL (Egypt), Rapporteur

The main items on the agenda were the following : the implementation of narcotics treaties and international control; the illicit traffic; the proposed Single Convention on narcotic drugs; the request of Afghanistan to be recognized as a State producing opium for export; the problems of drug addiction, coca leaf, cannabis, synthetic narcotic drugs; and scientific research on opium. The Commission decided to defer three questions until its twelfth session—the problem of barbiturates, the problem of khat, and the establishment of a United Nations Middle East Anti-Narcotics Bureau.

Representatives of Afghanistan, the Federal Republic of Germany, Hungary, Israel, Italy, Japan, Lebanon, the Netherlands, Pakistan, Portugal, Switzerland and Thailand participated in the consideration of items of particular concern to them.

Representatives of the World Health Organization, the Permanent Central Opium Board, Drug Supervisory Body, Permanent Anti-Narcotics Bureau of the League of Arab States, International Criminal Police Organization, World Federation of the United Nations Associations, and the World Union of Catholic Women's Organizations also took part in the work of the Commission.

The Commission took a number of decisions, the principal of which are mentioned below, and recommended resolutions to the Economic and Social Council dealing with the implementation of the narcotics treaties, technical assistance for narcotics control in general, and technical assistance to Iran in particular, which are noted under the next heading.

Illicit Traffic

In accordance with the established procedure, the Committee on Illicit Traffic, consisting of a nucleous of eight members of the Commission and with the attendance of four other members undertook a review of all the available material relating to the illicit traffic, basing itself upon the work of its Committee.

The Commission noted that opium and its derivatives, in particular morphine and diacetylmorphine, continued to be by far the most important drugs seized in the internal and international illicit traffic. There was no apparent decline in the traffic in raw opium, which, as in the past, seemed to be concentrated in the Far East and the Near and Middle East. A particularly disturbing feature of this traffic was the movement of opium from the interior of Asia through Thailand. The tendency commented upon by the Commission last year, for conversion into morphine and diacetylmorphine to take place nearer the source of cultivation of the opium, appeared to be increasing.

The Commission felt grave concern at the striking increase in seizures of morphine base and crude morphine hydrochloride; the most significant feature of the traffic was the existence of clandestine manufacture in both the Far East and the Near and Middle East. The Commission observed with serious misgiving that seizures of diacetylmorphine had also significantly increased and that clandestine manufacturing activities had been reported by several countries. North America continued to be the destination of much of the more highly organized traffic in this drug.

Seizures of prepared opium were significant mainly in the Far East and the Near and Middle East. Besides being a natural sequel to the traffic in raw opium in these regions, it indicated the existence of a deep-rooted habit despite the legal prohibition of opium smoking almost without exception in these regions.

Although the long-term decline in the quantities of cocaine seized throughout the world continued, it was observed that there was a persistent though small traffic in this drug in the Far East, Near and Middle East, Europe and North America.

The Commission observed that consumers of cannabis numbered millions in the world and that geographically it was the most widespread drug of addiction. A large proportion of this traffic consisted of indigenous production and consumption, largely of the less concentrated forms. International illicit traffic was likewise to a large extent between countries with common frontiers and was generally not very highly organized. There was also a widespread though small-scale traffic carried on largely by merchant seamen. However, there were some important exceptions to this general pattern; the traffic in hashish in the Near and Middle East, the traffic in charas in the Indo-Pakistan sub-continent which appeared to be declining, and the traffic in marihuana to the United States by overland and sea routes.

Some seizures of very small quantities of synthetic drugs, particularly in Europe, were noted. However, appreciable seizures of dimethylthiambutene were reported from Japan, and the Commission was informed that these seizures had their origin in stocks concealed prior to the extension of national control to this drug. The Commission also noted that there had occurred diversions of small quantities of synthetic drugs from licit sources of the same kind as have long been known to exist in respect of natural drugs.

The Commission specially reviewed the illicit traffic situation in the Lebanon and Thailand and the opium situation in Iran. It expressed its sympathy with the difficult problems of control facing the Lebanese and Thai authorities and expressed its appreciation of their efforts in combating the illicit traffic. It decided that the situations in the Middle East and the Far East were to a large extent regional problems involving also other countries in these regions, and requested that technical representatives from these other countries should participate in the relevant debates at future sessions. With

regard to Iran, the Commission took note of the recent information regarding opium addiction in that country and of the law totally banning the cultivation of the opium poppy and use of opium in Iran. It expressed its appreciation of the bold step taken and decided to keep a close watch on developments in that country.

Finally, the Commission made certain recommendations to assist the fight against illicit traffic. It drew attention to the need (*a*) for severe penalties for traffickers; (*b*) for close liaison between the different services of national administrations entrusted with the control and supervision of drugs; (*c*) for better exchange of information relating to the illicit traffic, both directly between governments and through the intermediary of international agencies; (*d*) for better co-operation in the matter of origin of drugs seized in the illicit traffic, and in this connexion to send samples of seized opium to the Secretary-General so as to use modern laboratory techniques in the determination of origin.

Diacetylmorphine

The Commission noted that all but fourteen countries had either prohibited the manufacture, import, export and in some cases the use of diacetylmorphine, or adopted a policy of prohibition. The majority of representatives on the Commission expressed the opinion that that drug could be replaced by other less dangerous drugs, but the view was also expressed that it was still irreplaceable especially for the treatment of incurable diseases.

Cannabis

The Commission noted that a number of countries where cannabis is grown for industrial purposes reported that there was no diversion of cannabis from the licit fibre or seed crops. Experiments were being undertaken in some countries to breed a narcotic-free variety of cannabis.

Coca Leaf

The Commission thanked the Government of Peru for the measures taken in order to control the production of the coca leaf, to abolish gradually the habit of coca leaf chewing, and to develop suitable programmes of health education among the population concerned. It decided to invite observers of the countries in which coca leaf is grown or chewed (Argentina, Bolivia, Chile, Colombia, Ecuador and Indonesia) to participate in its discussion of the coca leaf question at its twelfth session.

Synthetic Narcotic Drugs

The Commission asked the secretariat to prepare a general study of the world situation as regards synthetic narcotics and adopted a resolution in which, *inter alia*, it recommended governments to prohibit, except for medical or scientific research, the production, distribution and use of such new narcotic drugs as, in their opinion, were not indispensable to public health. It urged governments to encourage scientists to continue their research for analgesics without addiction-producing properties, and to pay special attention to the emergence of new narcotic drugs to ensure that no new drug which was or might be addition-producing should be placed on sale without control, even though the World Health Organization might not have pronounced upon its addiction-producing quality. It invited governments to make the medical and allied professions aware of any new narcotic drugs especially dangerous to public health that might be placed on the market.

Amphetamines

The Commission adopted a resolution noting the dangers arising from the abuse of amphetamines and recommended that governments should provide adequate measures of control for the prevention of such abuse.

Drug Addiction

The Commission noted that there were millions of addicts to opium, cannabis and coca leaf, and hundreds of thousands of addicts to manufactured drugs, and that drug addiction represented a very serious problem in a number of countries, especially where the illicit traffic in narcotics was an important factor in organized crime. There was generally no marked improvement in the situation; on the contrary, morphine and heroin addiction seemed to be increasing throughout the world. The Commission requested the Secretariat to prepare a general paper on drug addiction for its twelfth session.

Twenty-second session of the Economic and Social Council

At its twenty-second session, held in Geneva from 9 July to 9 August 1956, the Council considered and took note of the report of the Commission on Narcotic Drugs on its eleventh session.

The Council invited the Government of the Federal Republic of Germany to adhere to the Paris Protocol of 19 November 1948; those States eligible to become parties to the Opium Protocol of 1953 which had not already ratified or acceded to that Protocol to do so immediately so that it would enter into force as soon as possible; and the governments to communicate promptly their national laws and regulations relating to the control of narcotic drugs. (The Paris Protocol provides machinery for placing synthetic narcotic drugs under international control. The Protocol of 1953 provides for limiting the production of opium to medical and scientific needs and for the abolition of opium smoking and opium eating.)

Technical Assistance for Narcotics Control

The Council considered this subject on the initiative of the Commission on Narcotic Drugs and agreed that it was highly desirable that technical assistance should be provided in this field. It adopted a resolution in which it drew the attention of those concerned to the areas where such assistance might be expected to do the most good, indicated the types of technical services available, and specified the development of substitute crops as one of the forms of assistance that might be requested. It recommended that the United Nations and the specialized agencies give due consideration to requests for technical assistance in developing appropriate administrative, social or economic measures for dealing with problems raised by the illicit production of and traffic in narcotic drugs or by drug addiction. It requested the Secretary-General to investigate whether non-governmental agencies, such as foundations and universities, might give assistance in their fields of interest, and to report on offers of assistance received, and also on the extent to which it had been found possible to meet requests for technical assistance in narcotics control under existing resolutions.

Technical Assistance to Iran

Representatives on the Council stressed the importance of the step taken by Iran in banning the cultivation of the opium poppy, and sympathized with the difficulties arising from it. On the initiative of the Commission it considered the question of technical assistance requested by Iran to meet these difficulties. It adopted a resolution recommending the Government of Iran to submit special requests for such technical assistance as it considered necessary in connexion with the prohibition of poppy cultivation and with the suppression of opium addiction. It invited the technical assistance authorities to give due consideration to those requests, drawing their attention to the importance of the Iranian measures for the economic and social development of that country. The Secretary-General was asked to report in 1957 on the extent to which it had been found possible to meet those requests. The Council was informed that the technical assistance mission of the Food and Agriculture Organization in Iran was already assisting in the development of substitute crops, and that an *ad hoc* committee had been established for that purpose.

Draft Single Convention on Narcotic Drugs

The Council decided to request the Commission to study the second draft of the Single Convention at its twelfth session, and if necessary at its thirteenth session. It decided also that the second draft should be transmitted for comment to all governments represented on the Commission and the Council, and to all governments whose representatives took part as observers in the Commission's work on the Single Convention.*

Claim of Afghanistan to be included among Countries which may produce Opium for Export

The Commission has adopted a resolution recognizing the justice of the claim made by Afghanistan that it should be included among the countries authorized to produce opium for export. It had requested the Secretary-General to amend the second draft of the Single Convention accordingly. Several representatives on the Council expressed the fear that the success of the Iranian programme of prohibiting poppy cultivation and suppressing opium addiction might be jeopardized if Afghanistan were authorized to produce opium for export. The Council, considering that there were technical points that needed more consideration, adopted a resolution inviting the Commission to consider this matter further in the light of the Council's discussions.

Meeting of Scientists on Opium Research

The Council agreed with the Commission that a meeting of experts to review and evaluate the research carried out at the national and the international level to develop methods for determining the geographical origin of opium seized in the illicit traffic would serve a useful purpose. It decided that such a meeting of nine experts should be held in 1958.

Location of the Next Session of the Commission on Narcotic Drugs

The Council approved the calendar of conferences for 1957, which provides that the next session of the Commission on Narcotic Drugs will be held at Headquarters in New York from 29 April to 31 May 1957.

The Committee on Illicit Traffic will meet three working days in advance of the Commission.

* The Single Convention is prepared to replace all multilateral narcotics treaties (see in *Bulletin*, Vol. VII, No. 1, the article of H. L. Moy : The Single Convention on narcotic drugs; comments and possibilities).